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Sequential Sampling for Larvae of the Winter Moth, *Operophtera brumata* (Linn.). (Lepidoptera: Geometridae)[1]

By W. A. Reeks[2]

Introduction

The Forest Insect Survey of the Maritime Provinces has been observing an outbreak of the winter moth, *Operophtera brumata* (Linn.), since its discovery in Nova Scotia in 1949. In addition to noting annual changes in distribution, the Survey has made population counts on 8 to 20 tagged trees of red oak, *Quercus rubra* L., at each of seven locations along the south shore of the Province. These trees were sampled each year to show the abundance of larvae of the winter moth and the fall cankerworm, *Alsophila pometaria* (Harr.). Both species are commonly found on the same trees, and their life histories are similar. The collections and subsequent rearings also showed the degree of parasitism by native or introduced species of parasites. The defoliation of the trees was estimated at the time of sampling and again at the end of the feeding period to determine the relationship between larval population and defoliation. Some of the sampling data, which were collected from 1952 to 1955, are used to develop the sequential plan of sampling described herein. Use of the plan at the sampling stations should reduce the number of samples while retaining accuracy within predetermined limits.

The principles of sequential sampling have been described in a number of papers (2, 3, 6, 7, 9). These will not be reviewed in detail, as the writer is concerned only with the application of the method to sampling for winter moth larvae.

Sampling Method

The general procedure was described in an earlier paper (5), and a similar procedure is now being used for studying populations of insects attacking poplar in British Columbia (1). Preliminary studies of the distribution of winter moth larvae (5) showed that the inter-tree variation constituted the principal source of variance in population. There were no significant differences in population between quadrants or levels within the crown. Therefore, the universe for sampling should be considered a universe of trees rather than a universe of foliage without regard to the particular tree on which it occurs. The sample unit can be either the leaf or leaf cluster (the leaves on a current shoot). Henson (1) mentions leaf size as being an important source of variation, and suggests relating insect populations to leaf area. However, it is not possible to measure leaf area when the trees are severely defoliated.

Several factors should be considered before accepting a sampling unit. Repeated defoliation may cause a reduction in the number of leaves per leaf cluster, as well as the number of leaf clusters per branch or tree. The average defoliation of sample trees for the years 1953, 1954, and 1955 on one plot was 78 per cent, 21 per cent, and 45 per cent, respectively; the corresponding number of leaves per cluster was 6.9, 5.6, and 5.2. The number of leaf clusters per branch in the lower crown was noted for 10 tagged branches of severely

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defoliated trees in 1954 and 1955. The number of leaf clusters per branch averaged 26 both years, but it is not known what degree of increase in the number of leaf clusters could be expected each year in the absence of defoliation. There is also some evidence that repeated defoliation reduces leaf size.

The objects in sampling might influence the choice of the leaf or leaf cluster as a sampling unit. In ecological studies for the development of life tables, it would be necessary to determine the "absolute population" (4), which in case of the winter moth likely would be the population per tree. This would involve knowing the foliage production as well as the insect population each year. The foliage production probably would be expressed as total leaves per tree (or possibly numbers per acre, or leaf area if defoliation is light), and there would be no particular advantage in knowing the number of leaf clusters per tree. However, for sampling by the sequential method there are certain advantages in adopting the leaf cluster as the sampling unit. A reduction in the number of leaves per shoot from one year to the next would indicate an apparent increase in population per leaf, but not in population per leaf cluster (5). The leaf cluster can be sampled almost as quickly as the leaf, and it provides a larger sample of the population for rearings or other incidental data. Furthermore, the sequential method does not require an expression of absolute population. Instead, it is based on acceptable infestation classes.

Preliminary sampling (5) was used in developing different combinations of leaf clusters per tree that will provide equal reliability. For example, a sample of 12 leaf clusters from each of 8 trees was shown to be reliable as a fixed sample size. Therefore, this unit is used for the present sequential plan. Because of the low intra-tree variation in population, the leaf cluster may be taken from any part of the crown of the sample tree. Supplementary information, such as defoliation of the tree or leaf cluster, leaf size, or shoot length may also be noted in accordance with the objects of the sampling.

Since the larval populations of the winter moth in the sample areas were always over 92 per cent of the combined populations of the winter moth and cankerworm, the two species are combined in the population counts that follow.

The best sampling time for larval surveys is during the period when the majority of the larvae are in the fourth instar. This may be from about June 7 to June 15 along the south shore of Nova Scotia, depending on the season, but sampling during the first two weeks of June should generally be satisfactory.

Infestation Classes

Although the sequential plan is concerned with larval sampling on oak trees, it is desirable to indicate the degree of defoliation that can be expected from various levels of larval populations. The infestation classes (Table I) are determined from observations on individual trees for 2 to 4 consecutive years.

TABLE I

Infestation classes of winter moth based on larval numbers and defoliation of red oak trees

Infestation	Total larvae per tree-sample of 12 leaf clusters	Approximate percentage of defoliation at end of feeding period
Light	0–12	0–25
Moderate	15–36	35–80
Severe	42+	90–100

Sustained attacks of the winter moth may necessitate changes in the above interpretation, and the classification may not be acceptable in very recent infestations where the leaves tend to be larger. Reduction in leaf size or number can change the relationship between population and defoliation, but the classification is sufficiently broad that the relationship would have to change considerably before a revision of the classification becomes necessary.

Thompson (8), quoting work by Varley, reports cases of winter moth eggs hatching before the buds of host trees have burst. This forces the larvae to feed on the buds, some of which may die. Hence the foliage arising from the remaining buds may support only a relatively light larval population, and the true relationship between population and defoliation (and bud-killing) may be lost.

Sequential Plan

As shown by Morris (3), the first step in the application of the sequential plan is to establish the nature of the frequency distribution of the insect counts. Waters (8) states that forest insect survey data will generally fit one of the four following distributions, binomial, negative binomial, Poisson, or normal.

In the winter moth studies, the uniform defoliation within the tree crown and the solitary feeding habits of the larvae suggested some form of random distribution. This was confirmed by establishing the relationship between mean and variance for the larval count per leaf cluster, using a total of 170 observations on individual trees, which supported a wide range of population means. The Poisson distribution specifies that the mean and variance are equal. The distribution for the winter moth, although not strictly Poisson, can be treated by methods appropriate to the Poisson distribution.

Two kinds of errors are involved in sequential sampling (8), the risk α of calling a light infestation moderate or a moderate infestation severe and the risk β of calling a moderate infestation light or a severe infestation moderate. For sampling winter moth larvae α and β are set at 0.10.

The formulae for the Acceptance and Rejection lines (or Decision lines) are taken from Waters (8). These are shown for the light versus moderate infestation as follows:

$$d_1 = bn - h_1 \qquad \text{(lower line)}$$

$$d_2 = bn + h_2 \qquad \text{(upper line)}$$

where d is the cumulative number of larvae per tree-sample, n is the number of trees sampled, and the slope of the lines

$$b = \frac{m_2 - m_1}{\log_e m_2 - \log_e m_1} = 13.446$$

where $m_2 = 15$, $m_1 = 12$ (Table 1)

and the intercepts

$$h_1 = \frac{\log_e \frac{1-\alpha}{\beta}}{\log_e m_2 - \log_e m_1} = 9.847$$

and

$$h_2 = \frac{\log_e \frac{1-\beta}{\alpha}}{\log_e m_2 - \log_e m_1} = 9.847$$

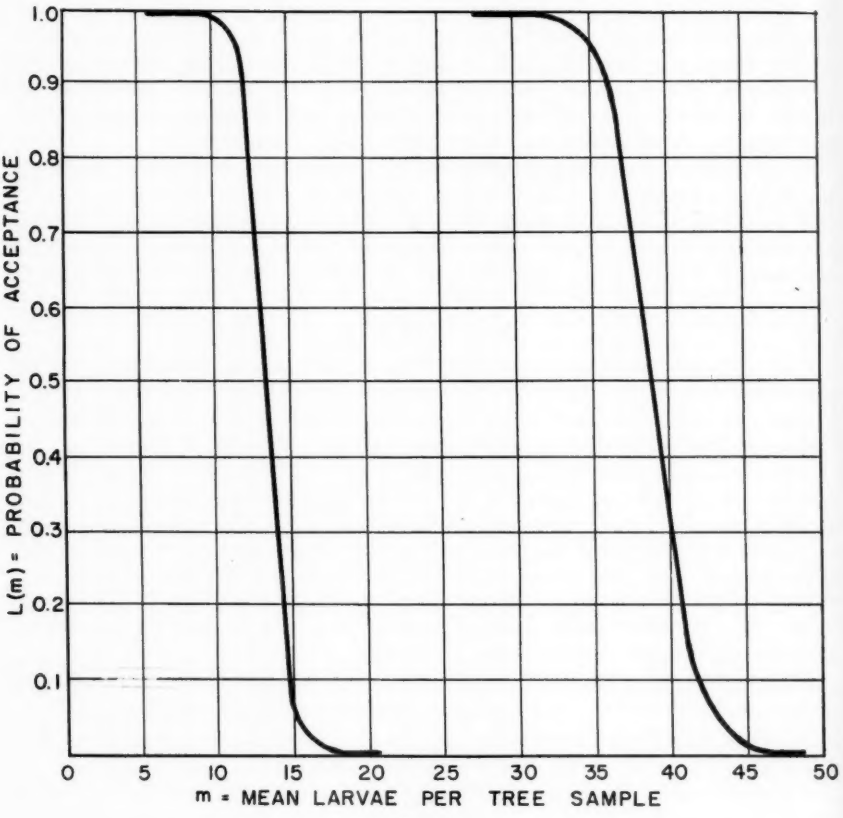

Fig. 1

Similarly, the Acceptance and Rejection lines for the moderate versus severe infestation can be drawn from the following data:

$b = 38.961$
$m_4 = 42$
$m_3 = 36$
$h_3 = h_4 = 14.266$

The Acceptance and Rejection lines can be shown graphically, but it is more convenient to tabulate the data for field use. This tabulation (Table II) shows that if sampling from one tree yields less than 3 or over 54 larvae there is no need to continue sampling. If the sample size falls within this range, sampling must be continued until the cumulative sample falls within the light, moderate, or severe zones.

Although not essential to the sequential plan of sampling, the Operating Characteristic curve (OC) and the Average Sample Number curve (ASN) help to show the usefulness of the plan. The OC curve gives the probability L(m) of a tree-sample m being accepted under the sequential plan. When the mean number of larvae per tree-sample m is 12, the probability L(m) of accepting h_1 (light infestation) is 0.9 (Fig. 1, left), and the probability of accepting h_2

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TABLE II

Cumulative number of winter moth larvae per tree-sample (12 leaf clusters per tree-sample) for one to ten sample trees. Based on values for d as shown in text.

Tree	Cumulative number of larvae per tree-sample				
		Light-moderate zone, continue sampling		Moderate-severe zone, continue sampling	
1	Light Zone	3 – 23	Moderate Zone	24 – 54	Severe Zone
2		17 – 37		63 – 93	
3		30 – 51		102 – 132	
4		44 – 64		141 – 171	
5		57 – 78		180 – 210	
6		70 – 91		219 – 248	
7		84 – 105		258 – 287	
8		97 – 118		297 – 325	
9		111 – 132		336 – 365	
10		124 – 145		375 – 404	

(moderate infestation) is 0.1. Similarly, at one level of population, m = 13.5, the probability of accepting h_1 and h_2 is about equal. The OC curve for the moderate versus severe infestation is to the right in Figure I. There is no overlapping of the two curves, indicating that rarely is there danger of calling a light infestation severe.

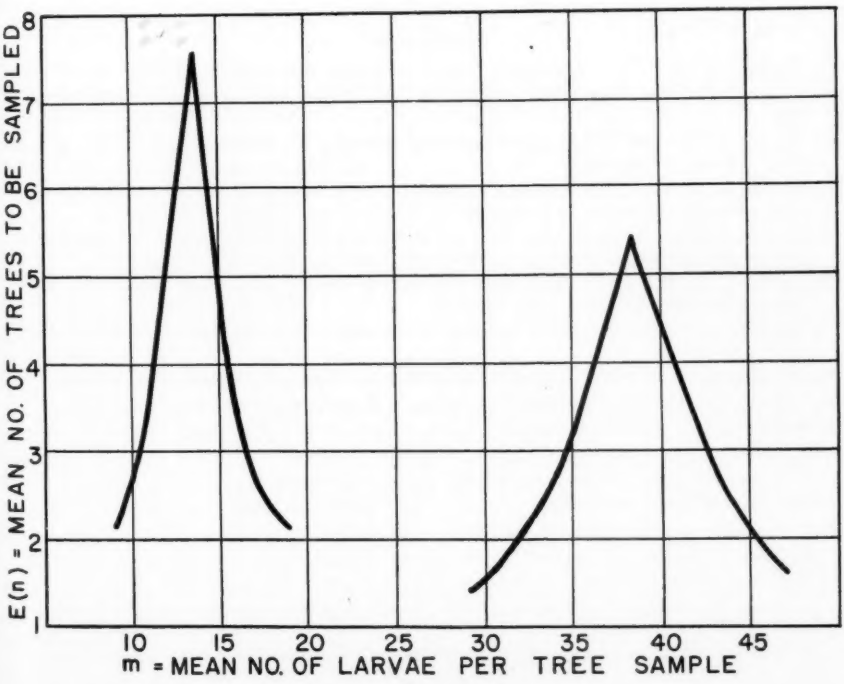

Fig. 2

The ASN curve shows the mean number of trees that probably would be sampled at various population levels (Fig. II). The highest degree of sampling is likely to be required when the mean number of larvae per tree-sample falls between 12 and 15 and between 36 and 41. Only rarely should it be necessary to sample more than 8 trees.

The formulae for the OC curve and the ASN curve are taken from Waters (8). The OC curve is calculated from

$$m = \frac{\frac{b}{h} \log_e \frac{L(m)}{1 - L(m)}}{\left(\frac{L(m)}{1 - L(m)}\right)^{\frac{1}{h}} - 1}$$, with b, h, and L(m) as defined above.

The ASN curve is calculated from E(n), number of trees sampled:

$$E(n) = \frac{L(m)\ (h_1 + h_2) - h_2}{b - m}$$

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References

(1) Henson, W. R. 1954. A sampling system for poplar insects. *Can. J. Zool.* 32: 421-433.

(2) Ives, W. G. 1954. Sequential sampling of insect populations. *Forestry Chron.* 30: 287-291.

(3) Morris, R. F. 1954. A sequential sampling technique for spruce budworm egg surveys. *Can. J. Zool.* 32: 302-313.

(4) Morris, R. F. 1955. The development of sampling techniques for forest insect defoliators, with particular reference to the spruce budworm. *Can. J. Zool* 33: 225-294.

(5) Morris, R. F. and W. A. Reeks. 1954. A larval population technique for the winter moth (*Operophtera brumata* (Linn.)) (Lepidoptera: Geometridae). *Can. Entomol.* 86: 433-438.

(6) Oakland, G. B. 1951. Sequential analysis. Processed publication, Biometrics Unit, Science Service, Can. Dept. Agr., Ottawa.

(7) Stark, R. W. 1952. Sequential sampling of the lodgepole needle miner. *Forestry Chron.* 28(2): 57-60.

(8) Thompson, G. H. 1954. Animal populations and forestry. *Nature.* 174: 813-815.

(9) Waters, W. E. 1955. Sequential sampling in forest insect surveys. *Forest Science* 1: 68-79.

New Species of Bark Beetles (Coleoptera: Scolytidae), Mostly Mexican. Part III[1]

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In this paper 12 new species and a new genus (*Prochramesus*) of scolytid beetles are described from Mexico, and one species is described from Honduras. Notes concerning geographical distributions of a few other Mexican species are included. The specimens taken in Mexico were all collected by the author while with the 1953 expedition of the Francis Huntington Snow Entomological Museum (University of Kansas, Lawrence). The new species represent the following genera: *Pseudohylesinus* (1), *Phrixisoma* (1), *Leperisinus* (1), *Phloeosinus* (1), *Chaetophloeus* (1), *Renocis* (1), *Phloeotribus* (1), *Prochramesus* (1), *and Chramesus* (4).

***Pseudohylesinus magnus*, new species**

This species is evidently more closely allied to *pullatus* Blackman than to other species of the genus, but differs in numerous respects including the following: body size much larger; frons flattened and more finely punctured below; pronotum more finely, closely punctured; strial punctures smaller; and the interstriae proportionately wider. A character of this species peculiar in the genus is the reticulate-granulate surface within each strial puncture.

Female.—Length 6.0 mm. (paratypes 5.5-6.1 mm.), 2.44 times as long as wide; body and scale colour black.

Frontal rectangle about equal in width and length; evenly convex above, transversely impressed above epistomal margin, flattened on a large triangular area above epistomal area, this triangular area divided on basal two-thirds by a narrow median carina and narrowly impressed on the median line from the carina to the apex of the triangle; triangular area finely, dorsal and lateral areas a little more coarsely punctured. Eye elongate-ovate, narrower below, entire; rather coarsely granulate. Antennal funicle seven-segmented; club conicle, the first segment about equal in length to the second segment.

Pronotum 1.15 times as wide as long; widest near base, posterior angles rather narrowly rounded, sides slightly arcuate and converging anteriorly, moderately constricted and transversely impressed behind the broadly rounded anterior margin; surface rather finely, very closely punctured, not granulate or asperate; middle of third median line slightly, narrowly elevated; vestiture consisting of inconspicuous short dark setae, with a few longer yellow hairs posteriorly.

Elytra wider than pronotum, 1.69 times as long as wide; the bases separately arcuate, crenulate, the overlapping marginal crenulations decreasing in size laterally; sides subparallel on basal two-thirds, rather narrowly rounded behind; striae impressed, rather narrow, the punctures moderately deep, very close, the surface within each puncture appearing rather coarsely reticulate-granulate; interstriae twice as wide as striae, nearly flat, transversely crenulate anteriorly, rather coarsely granulate-punctate on posterior three-fourths of disc (largely concealed by small, oval scales); vestiture consisting of abundant very dark brown to black scales, with small scattered patches (10-15 percent of surface) of light cinerous ones, and fine, short, hairlike, median, interstrial setae. De-

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clivity rather steep; striae and interspaces one and two slightly narrower than on disc; ninth interspace only slightly elevated, not serrate.

Male.—Unknown.

Type Locality.—Twenty-five miles west of Ciudad Hidalgo, Michoacan, Mexico.

Host.—*Abies* sp.

Type Material.—The female holotype and four paratypes were collected at the type locality on July 14, 1953. The holotype is in the Snow Entomological Collections, and the paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Phrixisoma minor*, new species**

Figs. 1, 2

Similar to *obesa* Blackman (types compared), but smaller in size, the interspaces narrower, the interstrial hairs much stouter with the median row proportionately longer, and three rows of granules on each interspace instead of four.

Male.—Length 1.7 mm. (paratypes 1.6-1.9 mm.), 2.0 times as long as wide; body colour black.

Frons convex, opaque, densely, finely granulate, with a fine, distinct, median carina on the lower half (absent in some paratypes), indistinctly impressed above the epistomal margin; vestiture fine, short, inconspicuous, slightly longer at epistomal margin. Eye completely divided, the halves rather broadly separated, the lower half slightly larger. Antennal scape long and slender; the funicle six-segmented; the club marked by three transversely arcuate sutures, the first partly septate.

Pronotum 1.33 times as wide as long, widest basally; posterior margin more or less bisinuate and rather sharply margined, the median area slightly produced; sides arcuately convergent toward the broadly rounded anterior margin; surface subopaque, rather finely, densely subgranulately punctured; vestiture fine, short.

Elytra 2.0 times as long as pronotum, about 1.3 times as long as wide, the bases finely crenulate; striae narrowly and deeply impressed, the punctures small, separated by a space about equal to their own diameters; interstriae about three times as wide as striae, flat on disc, more convex and narrower on declivity, rather finely and closely granulate, the granules arranged in three semi-definite ranks on posterior half of disc; pubescence consisting of short cinerous hairs, the median row on each interspace slightly longer on the posterior half.

Female.—Similar in size and habitus to the male, but with a sinuate line separating the pubescent and glabrous areas behind the eye (Figs. 1, 2), and with a corresponding anterolateral extension of the pronotum.

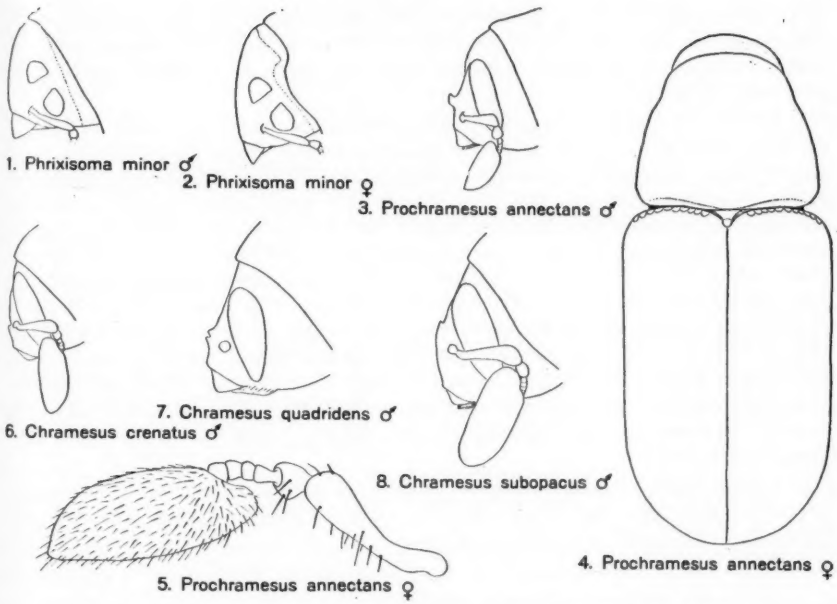
Type Locality.—LaCeiba, Honduras.

Host.—Unknown.

Type Material.—The male holotype was collected at the type locality on May 29, 1949; the female allotype and 12 paratypes were taken at the same locality on various dates between May 20, and August 26, 1949. All were taken at light by E. C. Becker. The holotype, allotype, and some paratypes are in the Canadian National Collection; other paratypes are in the collections of the U. S. National Museum.

***Leperisinus fasciatus* (Leconte)**

This species, described from Pennsylvania, is rather widely distributed in the eastern United States. A female specimen collected at Three Rivers, Texas, on June 27, 1938, by R. I. Sailer, and another female collected at San Juan


1. Phrixisoma minor ♂
2. Phrixisoma minor ♀
3. Prochramesus annectans ♂
6. Chramesus crenatus ♂
7. Chramesus quadridens ♂
8. Chramesus subopacus ♂
5. Prochramesus annectans ♀
4. Prochramesus annectans ♀

Figs. 1-2. Lateral aspect of head of *Phrixisoma minor*: 1, male; and 2, female.

Figs. 3-5. *Prochamesus annectans:* 3, lateral aspect of head of male; 4, dorsal aspect of female; and 5, antenna of female.

Figs. 6-8. Lateral aspect of head of male *Chramesus* spp.: 6, *crenatus;* 7, *quadridens;* and 8, *subopacus.*

del Rio, Durango, Mexico, on July 23, 1953, from a desert shrub, cannot be distinguished from female specimens taken near the type locality.

***Leperisinus mexicanus*, new species**

The two specimens assigned to this species have the colour, colour pattern, and many other features almost identical to those of *fasciatus;* however, they are distinctly larger, the striae are less strongly impressed (particularly on the declivity), the second declivital interspace is not as narrow, the frons of the male is much more shallowly concave, and the frontal carina of the female is less prominent.

Male.—Length 2.6 mm., 1.78 times as long as wide; colour piceous to black, with a variegated pattern of very dark brown (almost black) and white scales almost identical to that of *fasciatus.*

Frons very broadly and shallowly impressed, slightly concave, a finely raised median line indicating the presence of a carina on lower half; surface coarsely reticulate and sparsely, finely granulate-punctate particularly at sides and above; vestiture consisting of moderately abundant, rather long, coarse, white setae. Eye elongate-ovate, about 2.2 times as long as wide, entire, finely granulate. Antennal funicle seven-segmented; club distinctly flattened, about 1.9 times as long as wide, three transverse sutures indicated by rows of setae.

Pronotum 1.4 times as wide as long; widest at base, sides weakly, convergently arcuate, moderately constricted and transversely impressed just behind the nearly straight anterior margin; lateral areas with several rather large asperi-

ties; surface rather finely granulate-punctate, becoming coarse near summit; vestiture scalelike, dark brown in the asperate lateral and in the median posterior areas, white in the posterolateral areas, along the sides, and medially just behind the anterior margin.

Elytra 1.3 times as long as wide; wider than pronotum, sides subparallel on basal two-thirds, rather narrowly rounded behind; bases slightly raised, with rather large, overlapping crenulations from interspace one to eight; striae narrow, rather weakly impressed, the punctures rather small, deep, close; interstriae almost three times as wide as striae, nearly flat, the punctures rather fine, rugose; vestiture consisting of nearly erect, trilamellate scales, the median row longer, their colour very dark brown except for the small white patches located (1) just behind scutellum on interspaces one and two, (2) one-third the distance to declivity on interspaces six and seven, (3) at base of declivity on interspaces two to five, and (4) near apex of declivity; median row of setae on each interspace more slender and pointed anteriorly and on the sides. Declivity gradual; interstriae slightly narrower than on disc; sutural interspace moderately elevated; second interspace weakly impressed; striae slightly wider and less strongly impressed than on disc.

Female.—Similar to male except: length 3.0 mm; frons transversely impressed above epistoma, flat, with a weakly elevated median carina indicated just above the transverse epistomal impression.

Type Locality.—Tecamachaco, Puebla, Mexico.

Host.—An unidentified desert shrub.

Type Material.—The male holotype and female allotype were collected from the same gallery, at the type locality on July 2, 1953. They are in the Snow Entomological Collections.

***Phloeosinus taxodiicolens*, new species**

Closely allied to *taxodii* Blackman, but distinguished by the more finely punctured pronotum, the finer interstrial sculpture, the less strongly elevated first and third declivital interspaces, the more nearly scalelike declivital pubescence, and the more feebly impressed frons of the male.

Male.—Length 2.5 mm. (paratypes 2.4-2.6 mm.), 2.0 times as long as wide; body colour dark brown to almost black, with the elytra reddish brown.

Frons transversely flattened on lower half, convex above; rather coarsely granulate-punctate at sides and above, more finely punctured near centre; median carina broad, low; vestiture consisting of rather sparse yellow hair of median length. Eye about 3.2 times as long as wide, more than half divided by an emargination. Antennal club about 1.7 times as long as wide, sutures oblique.

Pronotum 1.18 times as wide as long; widest near base, sides arcuate, strongly constricted on anterior third, with a weak transverse impression behind the broadly rounded anterior margin; surface shining, closely and deeply, rather finely punctured; vestiture rather abundant, short, inconspicuous.

Elytra about 1.3 times as long as wide; wider than pronotum; sides subparallel on basal two-thirds, broadly rounded behind; striae narrow, strongly impressed, the punctures small, distinct, separated by one and one-half to two times their own diameters; interstriae on disc about three times as wide as striae, finely granulate-punctate, granules larger and uniseriate near declivity; setae rather numerous, stout. Declivity with interspaces slightly narrower than on disc; the second as wide as the third, flat, finely punctured, with one or two granules near apex; first and third convex and armed by rather coarse, black,

blunt serrations, crowded on third, sparse posteriorly on first; five to nine each with a few rather coarse serrations; vestiture rather short, coarse, almost scale-like.

Female.—Similar to the male in size and habitus; frons more convex, carina more strongly elevated above, with a weak impression above the carina; first and third declivital interstriae less strongly elevated, the serrations much smaller, and the second usually with a few serrations about equal in size to those on one and three.

Type Locality.—Tomazunchale, San Luis Potosi, Mexico.

Host.—Taxodium mucronatum.

Type Material.—The male holotype, female allotype, and 76 paratypes were collected at the type locality on June 21, 1953, from branches of the host tree; 29 other paratypes were taken twelve miles north of Rodeo, Durango, Mexico, on July 25, 1953. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Chaetophloeus lasius*, new species**

This species is similar to *hystrix* (Leconte), but differs as follows: elytra more finely and shallowly punctured, the striae not evident; third declivital interspace not armed; and the pubescence longer and more nearly plumose. The two species agree in antennal, frontal, and tibial characters, and in the general type of vestiture.

Female.—Length 2.4 mm. (paratypes 1.8-2.5 mm.), 1.88 times as long as wide; colour black, the pubescence yellowish- or brownish-white.

Frons with a rather strong, subconcave, transverse impression above epistoma; surface finely, rather sparsely punctured, largely concealed by rather abundant, long, plumose setae; a pair of widely spaced, pointed tubercles at upper level of eyes; mouthparts, epistoma, and bases of mandibles clothed with long yellow setae. Eye elongate-oval, entire; rather finely granulate. Antennal funicle five-segmented; club 1.3 times as long as wide, with three non-septate, slightly arcuate, transverse sutures, segment four almost equal to the combined length of segments one, two, and three.

Pronotum 1.33 times as wide as long; sides strongly, convergently arcuate, the anterior margin broadly rounded; surface uniformly, rather closely punctured, the punctures rather deep, moderately large; laterally armed by two paired groups of asperities, each group consisting of from one to four tubercles; vestiture long, moderately abundant, and conspicuously plumose.

Elytra 1.13 times as long as wide; the bases strongly elevated medially, each bearing about six marginal serrations that decrease in size laterally and three submarginal ones; striae not clearly evident, the surface covered with confused, close, moderately fine punctures, each puncture bearing a long, erect, more or less scalelike but evidently plumose seta. Declivity rather steep, sulcate, unarmed; striae scarcely evident; the setae in rows evidently corresponding to interspaces.

Male.—Frons more strongly impressed, and the antennal club more elongate than in the female.

Type Locality.—Seven miles southeast of Nochixtlan, Oaxaca, Mexico.

Host.—Unknown.

Type Material.—The female holotype, male allotype, and 62 paratypes were collected at the type locality on July 5, 1953, from an unidentied, resinous, sumac-like shrub. The holotype, allotype, and some paratypes are in the

Snow Entomolgical Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Renocis sulcatus*, new species**

Allied to *maclayi* (Bruck), but with the frons less strongly impressed, the frontal tubercles smaller, the lateral margins of the declivity more strongly elevated, and a distinctive colour pattern that includes a transverse band of dark brown scales across the base of the pronotum (largely white in *maclayi*).

Female.—Length 1.9 mm. (paratypes 1.7-2.1 mm.), 1.83 times as long as wide; body dark brown, the colour pattern of the vestiture variegated, consisting of dark brown and nearly white scales and hairlike setae.

Frons flat, a weak transverse impression above epistoma; epistomal margin with a short, sharp, median, pointed process and with a fringe of light yellow-cinerous setae; laterally armed by a pair of sharp, prominent tubercles at upper level of eyes; surface shining, finely punctured, largely concealed by the rather coarse, semi-erect, light brown and cinerous setae. Eye about 3.5 times as long as wide, the inner margin sinuate. Antennal club 2.5 times as long as wide, with three straight, aseptate sutures indicated by rows of setae.

Pronotum 1.5 times as wide as long, widest near base; basal margin broadly procurved; sides arcuate, strongly constricted and transversely impressed just behind the nearly straight anterior margin; surface rather finely, closely punctured, with interspaces minutely reticulate-granulate; two small groups of rather coarse asperities on each side; vestiture consisting of small, broad, recumbent scales, their colour dark brown on the median and basal areas, white in irregular lateral and anterior areas.

Elytra about 1.3 times as long as wide; wider than pronotum; sides subparallel on basal two-thirds, very broadly rounded behind; each basal margin elevated from first to fifth interspace and armed by seven coarse serrations that decrease in size laterally; striae weakly impressed, the punctures of moderate size and separated by a distance equal to their own diameters; interstriae about twice as wide as striae, the surface finely reticulate-granulate, finely punctured, and nearly concealed by numerous small, semi-erect, brownish and white scales, and median rows of longer erect scales; each elytron with a rather dense group of erect brown setae behind basal elevation; colour pattern irregular, small patches of white scales cover about forty to fifty percent of the surface. Declivity moderately steep, rather strongly sulcate between the third interspaces; the first striae more strongly impressed on upper half, the sutural interspaces very slightly elevated; third interspace rather high, with a rather prominent elevation bearing a group of erect scales at the junction with the seventh interspace, and with the longer median setae more numerous than on the disc and forming a partial double row.

Male.—Similar in size and habitus to female, but with the frons more strongly impressed, with the vestiture longer and more abundant; antennal club more slender; and setae on anterior margin of pronotum longer.

Type Locality.—Ixmiquilpan, Hidalgo, Mexico.

Host.—Unknown.

Type Material.—The female holotype, male allotype, and eighteen paratypes were collected at the type locality on June 23, 1953, from an unidentified roadside shrub. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; additional paratypes are in the Canadian National Collection and in the collections of the U. S. National Museum.

***Renocis eggersi*, new name**

The name *Renocis mexicanus* Eggers (1949, Ent. Blätt. 41-44: 149) is preoccupied (Blackman, 1940, Proc. U. S. Nat. Mus. 88: 397). It is proposed that Eggers' species be renamed in his honour.

***Phloeotribus pruni*, new species**

This species is very closely allied to *liminaris* Harris, but is distinguished by the more coarsely sculptured pronotum and elytra, by the presence of distinct interstrial granules, by the more numerous, more slender crenulations of the elytral bases, by the more shallowly impressed strial punctures, and by the coarser vestiture.

Female.—Length 2.1 mm. (paratypes 1.7-2.1 mm.), 2.1 times as long as wide; body colour brown.

Frons convex, with a slight transverse impression above epistoma, a feeble elevation between antennal bases, and a small shallow median fovea near centre; surface reticulate-granulate, with sparse shallow punctures; vestiture consisting of sparse yellow hair of medium length. Eye elontate-ovate, entire; finely granulate. Antenna as in *liminaris*.

Pronotum 1.17 times as wide as long; widest at base, sides weakly arcuate, feebly constricted just behind the broadly rounded anterior margin; surface reticulate, punctures of moderate size and density, shallow; vestiture fine, rather long and abundant.

Elytra 1.4 times as long as wide; sides subparallel on basal two-thirds, broadly rounded behind; basal margins separately arcuate and armed by about 15 rather narrow overlapping crenulations, the lateral ones sharply pointed; the submarginal crenulations smaller and more irregular than those on margin; striae not impressed, the punctures of moderate size, distinct but shallow; interstriae wider than striae, ornamented by two irregular ranks of small granules, except uniseriate on interspace one. Declivity rather steep, convex; striae narrower and a little more strongly impressed; interstrial granules uniseriate and more sharply pointed than on disc, only slightly larger on nine; none of the interspaces elevated. Vestiture consisting of erect, rather fine yellowish hair; the setae in a double row on each interspace of disc, except uniseriate on interspace one; stouter, but not longer than on declivity.

Male.—Similar to female except: epistomal area with a rather broad, very low, median elevation, the surface of the elevation conspicuously reticulate; the transverse impression above the epistoma obscure; and frons flattened, the median fovea obscure or absent.

Type Locality.—Fifteen miles northwest of Chihuahua, Chihuahua, Mexico.

Host.—*Prunus* sp.

Type Material.—The female holotype, male allotype, and 45 paratypes were collected at the type locality on July 29, 1953, from a native species of *Prunus* that grew along the bank of a stream. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Prochramesus*, new genus**

Closely allied to *Chramesus* Leconte, but differing as follows: body form slender (Fig. 4); pubescence essentially hairlike; antennal scape shorter, the club smaller and less strongly compressed; and the posterior margin of the pronotum not extended medially. This genus resembles *Chramesus* in having the antennal

funicle five segmented and attached to the side of the club; in having the club unsegmented and similar in shape (Fig. 5); and in having the frons of the male concave, with the lateral margins armed.

Description.—Frons flattened in the female, concave in the male; eye elongate-oval, entire; antennal scape rather long, but not extending beyond the posterior margin of the eye; funicle five-segmented and attached to the side of the moderately flattened, unsegmented club; pronotum only slightly wider than long, the posterior margin essentially straight, not extended medially, the sides not asperate; elytra elongate, the bases crenulate; vestiture consisting of hairlike and bristle-like setae, the short interstrial setae appearing almost plumose, but not scalelike.

Type Species.—*Prochramesus annectans*, new species.

***Prochramesus annectans*, new species**

Figs. 3, 4, 5

Female.—Length 2.3 mm. (paratypes 2.0-2.3 mm.), 2.33 times as long as wide, body colour black (including the antennal club).

Frons flattened on a more or less triangular area extending from epistoma to upper level of eyes, armed just above the level of the antennal insertion by a pair of prominent tubercles, the tubercles separated from one another by a space equal to half the distance between the eyes; vestiture consisting of a few short stout yellow setae. Eye elongate-oval, only slightly narrower ventrally, entire; rather finely granulate. Antennal scape extending to posterior margin of eye; funicle shorter than scape, ornamented by a few small scattered hairs; club rather large, tapered distally, similar to *Chramesus* spp. but less strongly flattened and with longer, less abundant vestiture.

Pronotum 1.12 times as wide as long; the posterior margin straight, the lateral margins slightly arcuate and converging anteriorly, rather broadly rounded in front, indistinctly impressed behind the anterior margin; surface of disc covered with small isolated granules with a few punctures interspersed, each granule and puncture giving rise to a short, yellow, bristlelike seta.

Elytra wider than pronotum, 1.69 times as long as wide; the bases separately arcuate, their margins elevated and armed by about eleven transverse crenulations; sides subparallel on more than the basal two-thirds, broadly rounded behind; surface shining; striae indistinctly impressed, the punctures moderately large, rather deeply impressed, separated by less than their own diameters; interstriae equal in width to striae, each with a median row of granulate punctures, the granules rather large and each bearing a moderately long bristle, additional short, rather broad setae in semi-definite rows arising from minute punctures between the strial punctures and the granules. Declivity steep, convex; the strial punctures and interstrial granules somewhat smaller than on disc, and the vestiture more conspicuous.

Male.—Similar to the female except: frons rather deeply, almost hemispherically concave below upper level of eyes; the excavation occupying only about half of the space between the eyes, its lateral margins not acute; the frontal tubercles larger, each laterally compressed and directed more dorsomedially than in the female and appearing subquadrangular from the lateral aspect (Fig. 3).

Type Locality.—Seven miles southeast of Nochixtlan, Oaxaca, Mexico.

Host.—*Nolena* spp.

Type Material.—The female holotype, male allotype, and 29 paratypes were collected at the type locality on July 5, 1953; 19 other paratypes were taken ten miles southeast of Oaxaca, Oaxaca, Mexico, on July 6, 1953. These beetles

were collected from the fruiting stalks of two species of *Nolena.* They were monogamous and constructed transverse galleries in the outer woody portion of the fruiting stalks. The galleries of the parent adults were usually transverse and similar to those of *Chramesus* species; those of the larvae were more or less straight and parallel to the axis of the stalk. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Chramesus subopacus* Schaeffer**

Fig. 8

This species was collected in Mexico at Encinal, Tamaulipas, on June 15, 1953 from *Condalia obtusifolia;* and at 18 miles west of Tehuantepec, Oaxaca, on July 8, 1953, from an unidentified shrub that grew in a hedge fence.

***Chramesus xylophagus*, new species**

Allied to *subopacus* Schaeffer, but the punctures of the striae and pronotum larger and deeper; the male frons not as deeply impressed and with lateral margins only slightly elevated, the small tooth located below the level of the antennal insertion (located as in *crenatus,* Fig. 6); and the female frons without a median fovea.

Male.—Length 1.9 mm. (paratypes 1.7-1.9 mm.), 1.6 times as long as wide; body colour dark brown to black.

Frons broadly, concavely impressed from epistoma to upper level of eyes, the lateral margins acutely, not strongly elevated and armed at lower level of antennal sockets by a pair of small blunt teeth (located as in *crenatus,* Fig. 6); the concavity sparsely clothed with rather small hairlike setae. Eyes and antennae similar to those of *subopacus.*

Pronotum 1.25 times as wide as long; widest posteriorly, the sides arcuately convergent anteriorly, slightly constricted just behind the broadly rounded anterior margin; posterior margin slightly produced medially as in related species; lateral areas very finely and sparsely asperate, median and posterior areas coarsely, rather shallowly punctured; episternal area finely reticulate-granulate, finely and very shallowly punctured; vestiture consisting of hair and stout bristles, not scalelike as in *subopacus.*

Elytra about equal in length and width, the sides subparallel on the basal half, very broadly rounded behind; anterior margin of each elytron armed by about twelve rather large overlapping crenulations, and a few submarginal ones, the marginal crenulations becoming narrower laterally; striae slightly impressed, the punctures moderately large and deep; interstriae about twice as wide as striae, each with a uniseriate row of granulate punctures; surface largely concealed by abundant, short, subplumose, scalelike, interstrial setae, and uniseriate rows of erect, stout, pointed, interstrial bristles, each bristle shorter than the distance between rows of bristles.

Female.—Similar to the male except: frons flattened, without an impression, elevated margins, or teeth; and pronotal asperities and the armature of elytral bases larger.

Type Locality.—Three miles southeast of Rosario, Sinaloa, Mexico.

Host.—An unidentified shrub.

Type Material.—The male holotype, female allotype, and ten paratypes were collected at the type locality on July 21, 1953, from a shrub that grew along a ditch at the edge of a cultivated field. The galleries were longitudinal, and

rather deep in the wood. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Chramesus quadridens*, new species**

Fig. 7

Allied to *xylophagus*, but the punctures of the lateral striae (seven to nine) larger and deeper, the proepisternum more nearly rugose and rather coarsely and deeply punctured, the male frons with a median fovea and with the lateral margins more strongly elevated and armed by two pairs of tubercles, and the female frons rugulose on the upper half.

Male.—Length 2.0 mm. (paratypes 1.8-2.1 mm.), about 1.6 times as long as wide; body colour dark brown to black.

Frons broadly, concavely impressed from epistoma to upper level of eyes, the lateral margins acutely elevated on lower two-thirds and armed at level of antennal sockets by two pair of blunt teeth of about equal size (Fig. 7); the concavity with a rather small, deep, median fovea just below the level of the upper end of the acute lateral margins; rather coarsely reticulate-granulate, except smooth just above epistoma; vestiture scanty, short. Eye and antenna similar to allied species.

Pronotum 1.36 times as wide as long; widest posteriorly, the sides arcuately converging anteriorly and slightly constricted just behind the broadly rounded anterior margin; anterior, lateral, and median areas rather finely asperate; posterior area coarsely, rather shallowly punctured, episternal area rugulose, and coarsely, rather deeply punctured; vestiture of disc consisting of stout bristles as in *xylophagus*.

Elytra 1.05 times as long as wide, the sides subparallel on basal half, very broadly rounded behind; anterior margin of each elytron armed by about eleven rather large crenulations that tend to overlap, and a few submarginal ones, the marginal crenulations becoming progressively narrower laterally; striae slightly impressed, the punctures moderately large and deep on the disc, becoming larger and deeper on the sides; interstriae about twice as wide as striae, each with a uniseriate row of granulate punctures; surface largely concealed by abundant, short, subplumose, scalelike, interstrial setae, and uniseriate rows of longer, erect, stout, blunt, interstrial bristles, each bristle shorter than the distance between rows of bristles.

Female.—Similar to the male except: frons flattened, without elevated margins, teeth, or median fovea; pronotal asperities and armature of elytral bases slightly larger.

Type Locality.—Seven miles southeast of Atlixco, Puebla, Mexico.

Host.—An unidentified leguminous vine.

Type Locality.—The male holotype, female allotype, and 15 paratypes were collected at the type locality on July 13, 1953. The type series, including larvae, was removed from the axis of stems about one-fourth of an inch in diameter. The holotype, allotype, and paratypes are in the Snow Entomological Collections.

***Chramesus striatus*, new species**

Closely allied to *asperatus* Schaeffer, but the punctures on the posteromedian area of the pronotum are not as large or as deep, the elytral bristles are much finer and shorter, the proepisternal areas are more coarsely and deeply punctured, the proepimeral areas are dorsoventrally rather coarsely striate, and the male frons is less deeply impressed.

Male.—Length 1.6 mm. (paratypes 1.4-1.8 mm.), 1.6 times as long as wide; body colour dark brown to black.

Frons rather broadly, deeply concave from epistoma to above the upper level of the eyes; lateral margins acutely, rather strongly elevated on the lower three-fourths and armed at lower level of antennal sockets by a pair of rather large, pointed teeth (located as in *crenatus*, Fig. 6); vestiture sparse, short. Eye and antenna as in allied species.

Pronotum 1.42 times as wide as long; widest near middle, slightly narrowed posteriorly; sides strongly arcuate, broadly rounded in front; finely asperate or granulate on anterior and lateral areas, a small posteromedian area rather finely, shallowly punctured; episternal area rather coarsely, deeply punctured; epimeral area with about five coarse, dorsoventral striations; discal vestiture consisting of short, stout bristles.

Elytra 1.05 times as long as wide, the sides subparallel on basal half, very broadly rounded behind; anterior margin of each elytron armed by about eight coarse crenulations, the lateral ones narrower; striae slightly impressed, the punctures moderately large and deep; interstriae about twice as wide as striae, each uniseriately granulate-punctate; surface largely concealed by abundant, short, rather slender interstrial scales, and uniseriate rows of slender interstrial bristles, the length of each bristle about equal to the distance between rows of bristles.

Female.—Similar to male except: frons flattened, transversely impressed above epistoma, and with a small median impression above; and pronotal asperities and elytral crenulations slightly larger.

Type Locality.—Seven miles south of Atlixco, Puebla, Mexico.

Host.—Evidently *Acacia* sp.

Type Material.—The male holotype, female allotype, and twelve paratypes were collected at the type locality on July 2, 1953, from the pith region of twigs and small branches of the host tree. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Chramesus crenatus*, new species**

Fig. 6

This species is evidently more closely allied to *striatus* than to other species that have similar frontal characters, but it differs conspicuously in vestiture, in the number and arrangement of the crenulations on the elytral bases, and in the sculpture of the proepisternal and proepimeral areas.

Male.—Length 1.5 mm. (paratypes 1.4-1.6 mm.), 1.6 times as long as wide; body colour very dark brown.

Frons rather broadly, deeply concave from epistoma to above the upper level of the eyes; lateral margins acutely elevated on lower three-fourths and armed at lower level of antennal sockets by a pair of blunt teeth (Fig. 6); vestiture very sparse and short. Antennal club large, but slightly shorter than in *asperatus*.

Pronotum 1.35 times as wide as long; widest posteriorly, convergently arcuate anteriorly and broadly rounded in front; finely asperate in anterior and lateral areas, coarsely and shallowly punctured in posteromedian area; episternal and epimeral areas rather coarsely reticulate, a few fine punctures scarcely evident; discal vestiture mostly of short slender bristles, a few scalelike setae evident especially in lateral areas.

Elytra about 1.05 times as long as wide, the sides subparallel on basal half, rather narrowly rounded behind; anterior margin of each elytron armed by a row of about twelve rather low crenulations, the lateral eight or nine of them basally united, not overlapping; striae impressed, the punctures rather small, impressed; interstriae about twice as wide as striae, uniseriately granulate-punctate; surface largely concealed by abundant, short, broad, interstrial scales and uniseriate rows of slender bristles, each bristle as long as the distance between rows of bristles.

Female.—Similar to male except: frons flattened, transversely impressed above epistoma, and with a small impression (a fovea in some specimens) above; a weakly developed arching elevation extending between antennal sockets; and pronotal asperities and elytral crenulations slightly larger.

Type Locality.—Twenty miles northwest of Vera Cruz, Vera Cruz, Mexico.

Host.—A large unidentified tree.

Type Material.—The male holotype, female allotype, and 169 paratypes were collected at the type locality on June 30, 1953, from longitudinal galleries in the cambium region of branches one-half to two inches in diameter. These beetles were associated with *Scolytus dimidiatus* Chapuis. The holotype, allotype, and some paratypes are in the Snow Entomological Collections; other paratypes are in the Canadian National Collection and the collections of the U. S. National Museum.

***Chramesus chapuisii* Leconte**

This species was collected at Los Abritos (20 miles northeast of Ciudad del Mais), San Luis Potosi, Mexico, from a broken stump about three inches in diameter of what appeared to be *Robinia* sp. It is a widely distributed bark boring species in the eastern and southern United States, but has not been reported previously from Mexico.

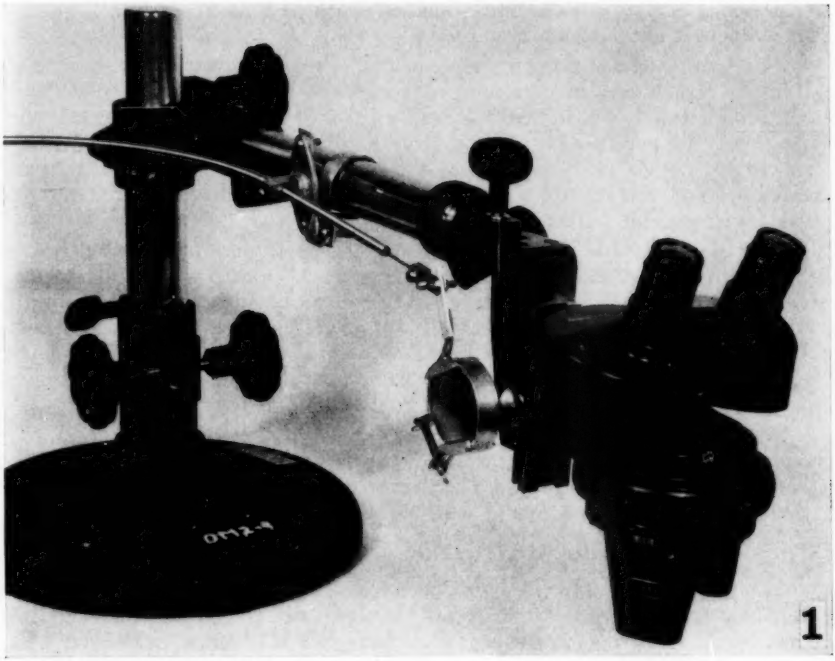
Flexible Cable for Foot Control of Microscope Focus[1]

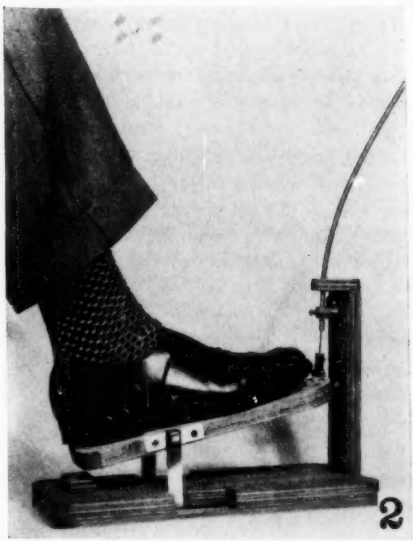
By D. W. Taylor and J. A. Chapman
Forest Biology Laboratory, Victoria, B.C.

When working with the stereoscopic microscope it is often desirable to use both hands for continuous manipulation of dissecting instruments or material under observation. In many instances, however, as when using the higher magnifications to study internal organs of insects, repeated focus adjustments are needed. The use of one hand for focus control is then a source of inefficiency and annoyance because it means frequent change of an instrument from its carefully adjusted position in the microscope field. Eyestrain and unsatisfactory performance follow attempts to reduce the number of adjustments made.

We have constructed and used a simple device for foot control of microscope focus. It is a logical extension of the method described by Mickey (1950) and also used by Chao (1955), and involves the coupling of a foot pedal with a focus knob lever by a flexible cable rather than a solid rod. Figures 1, 2, and 3 illustrate the essential features of this arrangement as adapted to two microscope models. We have found it to be effective and very helpful in work involving dissection of small insects. The method could be adapted for use with the

1Contribution No. 283, Forest Biology Division, Science Service, Department of Agriculture, Ottawa, Canada.





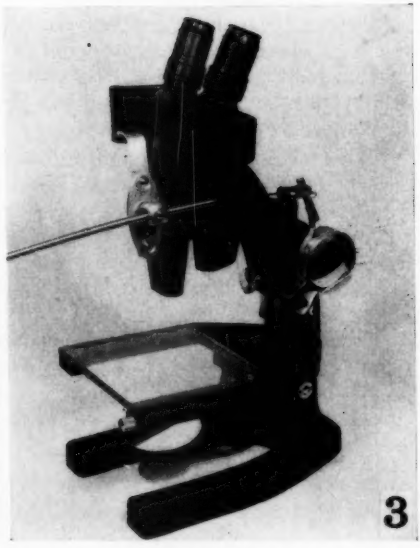


Fig. 1. Flexible cable attachment to horizontal arm type stand showing details of casing clamp, control wire socket and focus knob clamp.

Fig. 2. Plywood foot pedal control unit.

Fig. 3. Cable attachment to standard stereoscopic microscope stand. Casing clamp support is held in illuminator attachment socket.

(*All photographs by R. Banyard*)

compound microscope if the need should arise, as when both hands are required for continuous micromanipulator control.

For foot-pedal focus-knob coupling we have used Bowden wire and casing (¼ inch), which consists of a solid wire enclosed within a tightly coiled wire, both being flexible. A sufficient length of this or similar cable together with fittings may be purchased from any large hardware supplier for a few dollars. The fittings consist of casing clamps (swivel type), to hold the casing in the desired position, and control wire sockets, to fasten the inner wire to the movable parts of the control system.

The use of flexible cable permits ready change in position of either microscope or foot pedal and easy transfer of the work from one table or place to another. The stiffness of foot pedal movement can be adjusted by changing the radius of any of the bends in the cable. The rocker type pedal permits comfortable control of focus, and the extent of wire motion for a given foot movement will depend on the position of wire attachment in relation to the pedal fulcrum. This position could be made adjustable if desired.

Another way of changing the degree of control provided by given foot movements would be to alter the length of the focus knob lever. A suitable compromise between range and preciseness of focus control can be selected, depending on the requirements of work to be done. We have found that when the wire is attached two to three inches from the focus knob centre a satisfactory degree of control can be secured at magnifications up to 100 times with the foot pedal shown, and the range over which focus adjustment can be made will still be fairly satisfactory. It is possible, moreover, to allow for hand control at any time, thereby permitting ready change of this range. Optional hand control is desirable when examining objects which vary a great deal in size or in distance from the objectives, and may be secured by varying the tension of the lever clamp until some slipping of the focus knob can occur but sufficient friction remains for foot pedal control. Flanges or other means of keeping the clamp from gradually working off the focus knob are helpful with such an adjustment.

Foot focus control increases work effectiveness and ability to perform delicate and sustained manipulations using the stereoscopic microscope. Various modifications of the control system described could be made. We believe, however, that the simplicity of this method is such that anyone working with this type of microscope can use it. It requires a minimum of parts and construction, and no alterations in the microscope itself are needed.

References

Mickey, G. H. 1950. Foot focusing device for binocular microscope. *Stain Technology* 25: 217-218.

Chao, J. 1955. Sterile microdissection and isolation of malarial oocysts. *Science* 122 (3173): 763.

Aedes melanimon Dyar and Related Species[1]

By CHARLES S. RICHARDS, Scientist (R)

Aedes melanimon was described by Dyar (1924) from specimens collected at Bakersfield, California, as closely resembling *Aedes dorsalis* (Meigen) but differing in color, wing-scale pattern, and in the male genitalia. Freeborn (1926) considered *A. melanimon* to be an "aberrant form" or "race" of *A. dorsalis*.[2] Two species closely related to *A. dorsalis* are the American *Aedes campestris* Dyar and Knab, and *Aedes caspius* (Pallas) of Europe, Asia, and Africa.

A male mosquito collected in Nevada in 1953, the banded legs of which had been damaged, was identified by Dr. Harry D. Pratt, U.S. Public Health Service, and Dr. Alan Stone, U.S. National Museum, on the basis of the hypopygium as "nearest *Aedes klotsi* as compared with the type." Since the *dorsalis* pattern of leg banding was inconsistent with the description of *Aedes klotzi* Matheson (1933), a taxonomic discrepancy was indicated. This led the author to a study of *A. klotsi*, a species described from male and female adults collected at Fort Garland, Colorado. Upon further investigation, the Nevada male referred to above was determined as *A. melanimon*.

Studies concerning the status of *A. klotsi* and the comparison of larvae and adults of *A. melanimon* with those of *A. dorsalis* and related species, are presented in this paper. Grateful acknowledgment for invaluable aid is extended to Dr. Alan Stone for examining specimens and loaning material from the U.S. National Museum; Dr. Harry D. Pratt for identifications; Mr. James V. Smith for supplying reared specimens; associates at the Logan Field Station for advice, literature, and specimens; Dr. Don M. Rees and Mr. Lewis T. Nielsen of the University of Utah for the use of collections and literature; Mr. Barry D. Valentine for aid in collecting.

Methods

Larvae and adults of *A. melanimon*, *A. dorsalis*, *A. campestris*, and *A. caspius* were examined and compared. Numerous slides of larvae, male genitalia, and male and female tarsal claws (as employed by Vockeroth, 1950) were prepared from specimens collected in widespread localities. The material from the United States represented 13 western states, and included reared adults with associated larval exuviae of *A. melanimon* and *A. dorsalis*, and adults and associated larvae of *A. campestris*. Slides of female tarsal claws and male genitalia were made from topotypes of *A. melanimon* taken in the same year and by the same collector as the type specimen. Specimens of *A. caspius* from France, Italy, Algeria, Libya, Saudi Arabia, and Iran were examined. Type specimens of *A. melanimon*, *A. klotsi*, and several species now synonymized with *A. dorsalis*, were compared with *A. dorsalis* by Dr. Alan Stone.

Description of *Aedes (Ochlerotatus) melanimon* Dyar

Aedes melanimon Dyar, Ins. Ins. Mens. 12: 126, 1924.

Aedes dorsalis melanimon, Freeborn, Univ. Calif. Publ. Entom. 3: 368, 1926.

Aedes klotsi Matheson, Proc. Ent. Soc. Wash., 35: 69, 1935. *New Synonomy*.

Female. Head: proboscis black, occasionally with scattered light scales; palpi black, sometimes with a few white scales; tori with white scales on medial side; broad median area of occiput with narrow, whitish, curved scales and many erect, pale, forked scales; a submedian patch of brown scales and erect, forked,

[1]From the Communicable Disease Center, Public Health Service, U.S. Department of Health, Education, and Welfare, Logan, Utah.

[2]Since this manuscript was submitted for publication the following excellent and pertinent article has appeared: Barr, A. R., The resurrection of *Aedes melanimon* Dyar, Mosquito News, 15 (3): 170-172, 1955.

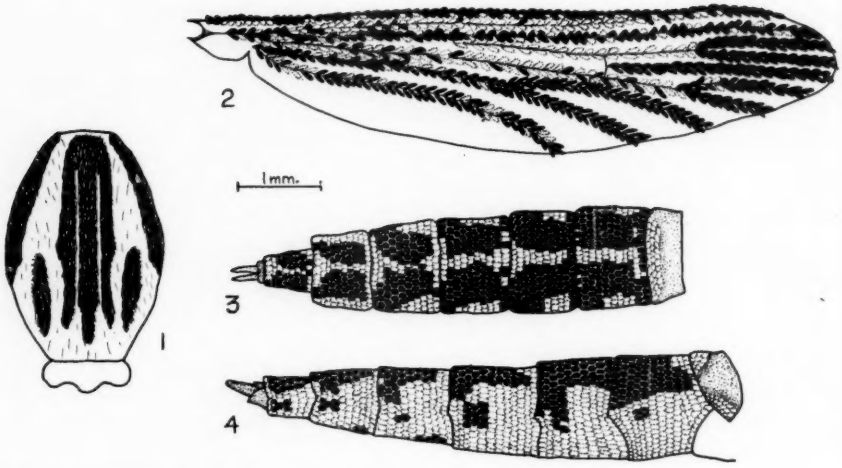


Fig. 1. *A. melanimon*, mesonotum, female.
Fig. 2. *A. melanimon*, wing, female.
Fig. 3. *A. melanimon*, dorsal view of abdomen, female.
Fig. 4. *A. melanimon*, lateral view of abdomen, female.

black scales bordering the median pale area on either side; lateral portion of occiput with broad, appressed scales, light brown to white sprinkled with black. Thorax: mesonotum (Fig. 1) with dark brown scales in a broad median band, separated from brown lateral areas by narrow lines of white scales; pleurae with scales broad and white; legs with white tarsal bands both basal and apical as in *A. dorsalis*, tarsal claw formula 1.18:0.82 (Fig. 5) with main claw straight in the middle and secondary claw relatively long and narrow; wing (Fig. 2) with costa dark-scaled along anterior margin except at base, otherwise occasionally with a few white scales; subcosta mixed as to scale color with light scales predominating; R-1, R-2, and R-3 mixed with dark scales predominating; R-4 + 5 all dark-scaled; M with light scales predominating except for distal ends which are dark-scaled; cubitus with dark scales predominating; anal mixed, with either dark or light scales predominating in various specimens. Abdomen (Figs. 3 and 4): dorsally with black scales predominating and extending laterally on the posterior half of most segments; seventh segment black except for a few white scales; dorsomedian white stripe sometimes incomplete, involving from two to seven segments in different specimens; ventral aspect white except for median black spots on segments 4-6.

Male. Hypopygium (Figs. 17-21): apical lobe large and bearing a few short, broad setae medially, almost bare on dorsal surface; basal lobe shelf-like, attached to basistyle posteriorly and not constricted at base, with outer spine short and close to the main long spine which is recurved at end, medial surface of lobe bearing many delicate setae; claspette stem curved, with filament longer than stem and widely expanded on the distal two-thirds. Mesonotum, pleural area, wings, and third tarsal claws similar to female.

Fourth-instar larvae. Head: upper and lower head hairs usually single, occasionally double. Thorax (Fig. 9): prothoracic hair formula 1(2),1,2-1-2(1,3),1,3; dorsal mesothoracic hair number one small, approximately the same length as the laterally adjacent hair, reaching about halfway to the head and usually two-

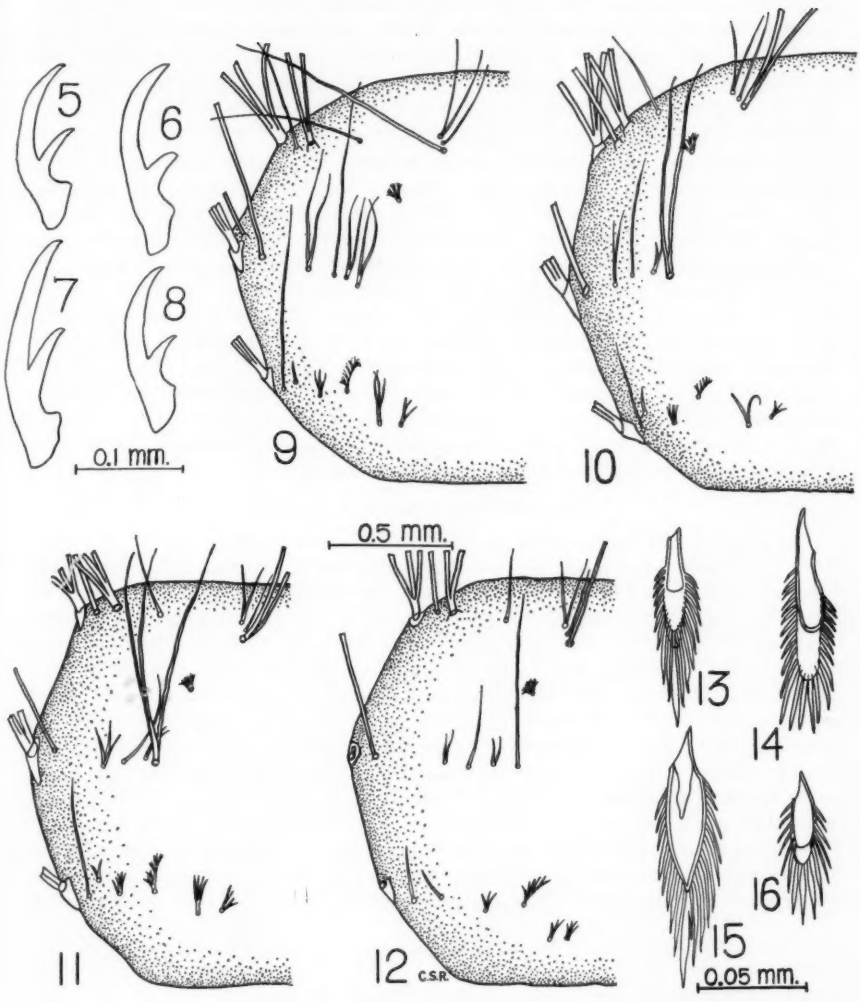


Fig. 5. *A. melanimon*, tarsal claw, female.
Fig. 6. *A. dorsalis*, tarsal claw, female.
Fig. 7. *A. campestris*, tarsal claw, female.
Fig. 8. *A. caspius*, tarsal claw, female.
Fig. 9. *A. melanimon*, dorsal view, left half of thorax, 4th instar larva.
Fig. 10. *A. dorsalis*, dorsal view, left half of thorax, 4th instar larva.
Fig. 11. *A. campestris*, dorsal view, left half of thorax, 4th instar larva.
Fig. 12. *A. caspius*, dorsal view, left half of thorax, 4th instar larva.
Fig. 13. *A. melanimon*, posterior comb scale, 4th instar larva.
Fig. 14. *A. dorsalis*, posterior comb scale, 4th instar larva.
Fig. 15. *A. campestris*, posterior comb scale, 4th instar larva.
Fig. 16. *A. caspius*, posterior comb scale, 4th instar larva.

branched. Abdomen: first four lateral abdominal hairs double (first two frequently, and third and fourth rarely, triple), fifth double, sixth single (rarely double); comb scales 16-27 (av. 21.9) in triangular patch and each with a prominent apical spine and smaller lateral spines (Fig. 13); siphon with 16-28 (av. 20.3) pecten teeth, ventral tuft of 4-8 branches located 52-60% (av. 55%) of siphon length from base; anal gills usually short and budlike.

Distribution. Specimens of *A. melanimon* have been examined from the following counties: California: Kern, Modoc, and Los Angeles; Colorado: Alamosa, Costilla, Delta, Mesa, Moffat, Rio Grande, Routt, and Weld; Montana: Blaine; Nebraska: Morill and Scottsbluff; Nevada: Churchill, Douglas, Elko, Eureka, Humboldt, Lander, Lyon, Mineral, Ormsby, Pershing, and Washoe; New Mexico: Sandoval and Valencia; Utah: Cache, Davis, Juab, Salt Lake, and San Juan; Washington: Benton and Franklin; Wyoming: Hot Springs. Specimens of *A. melanimon* from Idaho are included in U.S. National Museum collections. Specimens examined to date from Arizona, South Dakota, and Texas were all *A. dorsalis*. The material studied indicates that *A. melanimon* is relatively abundant in California, Colorado, Montana, Nebraska, and Nevada; rare in Utah. To date the number of specimens examined from the other states is not adequate to indicate the relative abundance of the two species.

Comparison with Related Species

In Table 1 and the illustrations (Figs. 1-33), various characters of *A. melanimon*, *A. dorsalis*, *A. campestris*, and *A. caspius* are compared. Although additional specimens were examined, the counts and measurements for larval characters in the table were based on: 25 specimens of *A. melanimon* from 7 widespread localities, 25 specimens of *A. dorsalis* from 9 localities, 12 specimens of *A. campestris* from 3 localities, and 12 specimens of *A. caspius* from 3 localities. For additional illustrations and discussions of *A. dorsalis*, *A. campestris*, and *A. caspius* see Barraud (1934), Manfridi (1953), Marshall (1938), Natvig (1948), and Rempel (1950, 1953). Although the mesonotal pattern of *A. dorsalis* is quite variable (Mail, 1934), *A. melanimon* has exhibited little variation in several thousand specimens examined. Although Dyar described the wings of *A. melanimon* as all dark-scaled or at least uni-colorous, no such specimens have been seen. Topotypes of *A. melanimon* taken by the original collector, as well as recent collections from near the type locality, have wings mostly dark-scaled, but close examination reveals a few white scales on several veins. White scales are pre-

Fig. 17. *A. melanimon*, dorsal view of basistyle of male hypopygium.
Fig. 18. *A. melanimon*, medial view of basistyle.
Fig. 19. *A. melanimon*, dorsal view of basal lobe of basistyle.
Fig. 20. *A. melanimon*, medial view of basal lobe.
Fig. 21. *A. melanimon*, lateral view of claspette of male hypopygium.
Fig. 22. *A. dorsalis*, dorsal view of basistyle of male hypopygium.
Fig. 23. *A. dorsalis*, medial view of basistyle.
Fig. 24. *A. dorsalis*, dorsal view of basal lobe of basistyle.
Fig. 25. *A. dorsalis*, lateral view of claspette of male hypopygium.
Fig. 26. *A. campestris*, dorsal view of basistyle of male hypopygium.
Fig. 27. *A. campestris*, medial view of basistyle.
Fig. 28. *A. campestris*, dorsal view of basal lobe of basistyle.
Fig. 29. *A. campestris*, lateral view of claspette of male hypopygium.
Fig. 30. *A. caspius*, dorsal view of basistyle of male hypopygium.
Fig. 31. *A. caspius*, medial view of basistyle.
Fig. 32. *A. caspius*, dorsal view of basal lobe of basistyle.
Fig. 33. *A. caspius*, lateral view of claspette of male hypopygium.

dominant on the subcosta and on most of the media as described above. It might be noted that the illustrations of hypopygia of "*A. dorsalis*" in Carpenter, *et al.* (1946) and Yamaguti and LaCasse (1951) appear to be of *A. melanimon.*

Series of *A. melanimon* males and females were compared with the types of *A. klotsi* by Dr. Alan Stone. The type material of *A. klotsi* consists of three males and two females. Although badly rubbed, the males show a faint indication of tarsal bands crossing the joints on some of the legs. One of the males shows the middorsal row of pale scales on the abdomen. The genitalia of all

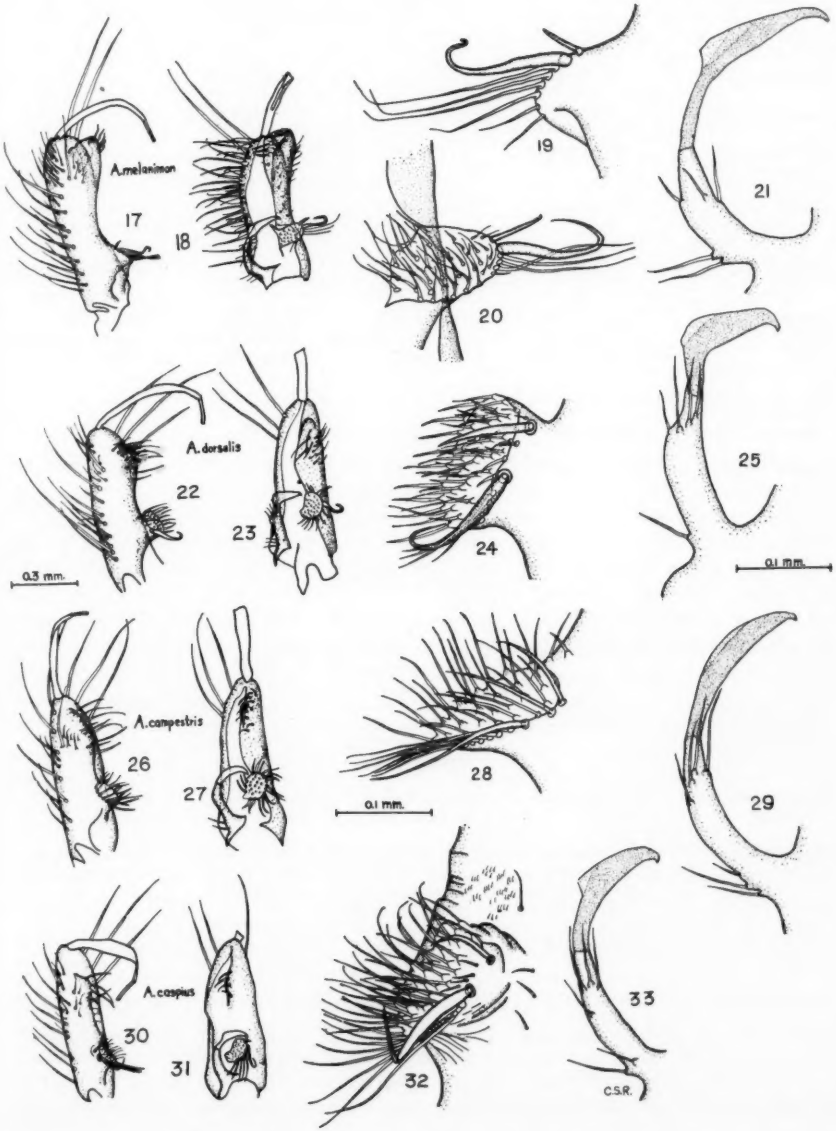


TABLE I

Comparison of Characters in *Aedes melanimon*, *Aedes dorsalis*, *Aedes campestris*, and *Aedes caspius*

Character	Aedes melanimon	Aedes dorsalis	Aedes campestris	Aedes caspius
FOURTH-INSTAR LARVA				
Upper head hair	1	1	3	1
Lower head hair	1	1	1	1
Teeth on mentum	19-25 (22)	17-23 (19)	17-19 (18)	18-21 (19)
Prothoracic hair formula	112-1-213	212-1-213	212-1-313	112-1-213
Dorsal, mesothoracic hair No. 1	2-branched, short	2-branched, long	3-branched, long	single, medium length
Lateral abdominal hairs	332221	332222	222222	222221
Comb scales	16-27 (22); prominent apical spine	18-34 (24); terminal spines subequal	24:29 (26); terminal spines subequal	18-34 (24); terminal spines subequal
Siphon tuft, percent of distance from base of siphon	52-60 (55)	43-56 (52)	56-61 (59)	51-62 (57)
Pecten teeth	16-28 (20)	14-22 (18)	20-25 (22)	16-26 (19)
FEMALE ADULT				
Pleural scales	broad, white	narrow, cream to straw	narrow, cream to straw	broad, white
Dark scales on dorsum of abdomen	extending laterally	two patches on each segment surrounded by light scales	two patches on each segment surrounded by light scales	extending laterally
7th abdominal segment	mostly dark-scales	mostly light-scaled	mostly light-scaled	dark and light, varied
Wing scales	dark predominate, concentrated on certain veins	light predominate, dark concentrated on certain veins	light predominate, dark scattered	light and dark, scattered
Anterior edge of costa	dark-scaled	mostly light-scaled	mostly light-scaled	light and dark scales mixed
Tarsal claws	formula; 1.18:0.82; primary claw straight in middle	formula: 1.15:0.85; primary claw evenly curved	formula: 1.3:0.7; primary claw almost straight	formula: 1.17:0.83; primary claw evenly curved

TABLE I—*Continued*

Character	Aedes melanimon	Aedes dorsalis	Aedes campestris	Aedes caspius
		MALE ADULT		
Apical lobe, hypopygium	prominent; almost bare dorsally	inconspicuous; dorsal setae numerous	inconspicuous; dorsal setae numerous	inconspicuous; almost bare dorsally
Basal lobe, hypopygium	shelf-like, attached to basistyle posteriorly, with very short spine close to large recurved spine, medial surface with delicate setae	constricted at base, with short spine separated from large recurved spine, medial surface with coarse setae	attached to basistyle posteriorly, usually with spines all similar, medial surface with coarse setae	attached to basistyle posteriorly, with short spine separated from large recurved spine, medial surface with coarse setae
Claspette of hypopygium	filament longer than curved stem, with slight angle and outer, angular flange	filament shorter than almost straight stem, with sharp angle and outer, angular flange	filament equal to curved stem, evenly curved and without angular flange	filament equal to almost straight stem, evenly curved and with outer, angular flange

three specimens agree very well with *A. melanimon*. The three males all show scales on the hypostigial area as in *A. melanimon* and *A. dorsalis*. The two females show no trace of banding on the legs; the one with scales on the abdomen shows distinct transverse bands but no median stripe; there are no pale scales on the hypostigial area. Dr. Stone concluded that the males (one of which was the holotype) described as *A. klotsi* are *A. melanimon*, and that the female paratypes of *A. klotsi* are not the same species, though what they are was not determined. It is of interest to note the similarity of the illustrations of male genitalia of *A. klotsi* and *A. dorsalis* (apparently *A. melanimon*) in Yamaguti and LaCasse (1951).

Dr. Stone examined the tarsal claws of type specimens of *curriei* Coquillett, *quaylei* Dyar and Knab, *lativittata* Coquillett, and *mediolineata* Ludlow, and found all to have claws shaped as in *A. dorsalis*, not as in *A. melanimon*.

Discussion

The distribution of the adults of *A. melanimon* and *A. dorsalis* suggests a difference in the preferred habitats of the larvae. *Aedes melanimon* apparently predominates in some areas of California and Colorado, *A. dorsalis* predominates in most of Utah, and the two species are both abundant in Nevada and Montana. In 1953, widespread collections were made in Nevada. In that part of the state included in the Great Basin, 1,379 adults of *A. melanimon* were taken in 76 collections, and 1,310 adults of *A. dorsalis* were taken in 65 collections. In collections from Davis County, Utah, in 1954, one specimen of *A. melanimon* was found, while 21,300 *A. dorsalis* were identified. *A. dorsalis* has been known to tolerate rather high salinity. If this tolerance does not apply equally to both species formerly considered under this name, it may help explain local differences

in distribution. Data show that larvae of both species are often associated with irrigation. Seasonal data suggest that *A. melanimon* as well as *A. dorsalis* may have several broods each year.

The differentiation of *A. dorsalis* and *A. melanimon* is of considerable importance. From available data it can be concluded that both are abundant throughout the western United States, but local distribution differs. *A. dorsalis* is known to occur in Canada and in Europe and Asia. Further knowledge of the distribution of *A. melanimon* must await differentiation of the two species wherever *A. dorsalis* is found.

Both *A. dorsalis* and *A. melanimon* attack man readily, inflicting painful bites, and are among the most important pest species in many areas because of their abundance. In addition to annoyance, the relationship of one or both of these species to certain arthropod-borne encephalitides is important.

Western Equine Encephalitis (WEE). The WEE virus has been recovered from wild specimens of *A. dorsalis* taken in California and Colorado. Two isolations were made by Hammon and Reeves (1945) from *A. dorsalis* collected in the San Joaquin Valley, California, in 1943. WEE virus was isolated from two lots of *A. dorsalis* from a total of 4,492 specimens collected in Weld County, Colorado, in 1950 (Thompson *et al.*, 1951). A total of 7,885 mosquitoes was tested. Of seven species, including 1,141 *Culex tarsalis* Coq., only *A. dorsalis* was found to be infected. The following quotation (*loc. cit.*, p. 290) is of interest:

> "Among the mosquitoes identified throughout the summer as *A. dorsalis* it was noted that considerable numbers of a dark form appeared which were readily distinguishable, morphologically, from the normal *A. dorsalis*. The dark form of *A. dorsalis* has also been noted in California and one of the 2 lots infected with WEE virus consisted entirely of the dark form. The dark form was routinely placed in ampoules separate from the normal *A. dorsalis*."

Five specimens from Weld County, Colorado, in the collection of the University of Utah have been examined. Four are *A. melanimon* and one is *A. dorsalis*. There is little doubt that the "dark form" found infected with WEE virus was *A. melanimon*.

St. Louis Encephalitis Virus. One isolation of this virus was made from *A. dorsalis* caught in Kern County, California (Hammon and Reeves, 1945).

California Encephalitis Virus. This virus was first isolated from a group of 80 *A. dorsalis* collected in Kern County, California, in 1943 (Hammon, *et al.*, 1952). These authors concluded that a strain isolated from 62 *A. dorsalis* collected in Kern County in 1944 and a strain isolated from 60 *C. tarsalis*, also from Kern County, were closely related or identical to the 1943 strain. It was demonstrated by Reeves and Hammon (1952) that *A. dorsalis* and three other mosquito species could be experimentally infected and would retain the virus for 7 to 8 days, also that *A. dorsalis* was able to transmit the virus to a rabbit in one of two experiments.

Since each of the above viruses was isolated from *A. dorsalis* from Kern County, California, it is of interest to note again that the type locality for *A. melanimon* is also in Kern County. The majority of a small series of larvae and adults from Kern County examined by the author were *A. melanimon*, Some of the reported isolations of virus encephalitides from *A. dorsalis* should probably be attributed to *A. melanimon*.

Conclusions

Comparison of *A. melanimon* with *A. dorsalis*, *A. campestris*, and *A. caspius* reveals differences distinct and consistent enough to conclude that four separate species exist. *Aedes melanimon* and *A. dorsalis* differ more from each other in many respects than either does from *A. caspius*. *A. melanimon* Dyar is considered a valid species and *A. klotsi* becomes a synonym of *A. melanimon*.

Summary

1. *Aedes melanimon* Dyar, found to be a valid species, is distinguished from *Aedes dorsalis* (Meigen), with which it has been confused, also from *Aedes campestris* Dyar and Knab, and *Aedes caspius* (Pallas). The larva of *A. melanimon* is described, and the adult is characterized more completely.

2. *Aedes klotsi* Matheson is shown to be a synonym of *Aedes melanimon*.

3. The distribution of *Aedes melanimon*, based on specimens examined to date by the author, includes nine of the Western States, in some of which it is one of the most abundant pest species.

4. Some of the *Aedes dorsalis* which have been reported as naturally infected with three of the virus encephalitides were in all probability *Aedes melanimon*.

References

Barraud, P. J. 1934. The fauna of British India. 463 pp. London.

Carpenter, S. J., W. W. Middlekauff, and R. W. Chamberlain. 1946. The mosquitoes of the southern United States east of Oklahoma and Texas. *Amer. Midland Nat. Monog.* No. 3, 292 pp.

Dyar, H. G. 1924. Two new mosquitoes from California. *Ins. Ins. Mens.* 12: 125-127.

Freeborn, S. B. 1926. The mosquitoes of California. *Univ. Calif. Publ. Entom.* 3: 333-460.

Hammon, W. McD., and W. C. Reeves. 1945. Recent advances in the epidemiology of the arthropod-borne encephalitides. *Am. J. Public Health* 35: 994-1004.

Hammon, W. McD., W. C. Reeves, and G. Sather. 1952. California encephalitis virus, a newly described agent. II. Isolations and attempts to identify and characterize the agent. *J. Immunol.* 69: 493-510.

Mail, G. A. 1934. The mosquitoes of Montana. *Mont. Agr. Exp. Sta. Bull.* 288, 72 pp.

Manfridi, F. 1953. Su alcune caratteristiche morfologiche dell' *A. (Ochlerotatus) caspius* Pallas. *Rendiconti Instituto Superiore di Sanita* 16: 204-210.

Marshall, J. F. 1938. The British mosquitoes. 341 pp. London.

Matheson, R. 1933. A new species of mosquito from Colorado. *Proc. Ent. Soc. Wash.* 35: 59-71.

Natvig, L. R. 1948. Contributions to the knowledge of the Danish and Fennoscandian mosquitoes. 567 pp. Oslo.

Reeves, W. C., and W. McD. Hammon. 1952. California encephalitis virus, a newly described agent. III. Mosquito infection and transmission. *J. Immunol.* 69: 511-514.

Rempel, J. G. 1950. A guide to the mosquito larvae of western Canada. *Canad. J. Research D.* 28: 207-248.

Rempel, J. G. 1953. The mosquitoes of Saskatchewan. *Canad. J. Zool* 31: 433-509.

Thompson, G. A., B. F. Howitt, R. Gorrie, and T. A. Cockburn. 1951. Encephalitis in Midwest. VI. Western equine encephalitis virus isolated from *Aedes dorsalis* Meigen. *Proc. Soc. Exper. Biol. and Med.* 78: 289-290.

Vockeroth, J. R. 1950. Specific characters in tarsal claws of some species of *Aedes*. *Can. Entomologist* 82: 160-162.

Yamaguti, S., and W. J. LaCasse. 1951. Mosquito fauna of North America. Office of the Surgeon, Hq. Japan Logistical Command, APO 343: Part V, Genus *Aedes*. 265 pp., 116 plates.

A Study of the Environmental Factors Affecting the Dispersion of House Flies (*Musca domestica* L.) in a Dairy Community near Fort Whyte, Manitoba[1]

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Introduction

House fly control as practiced at present requires a combination of thorough sanitation supplemented with the use of insecticides. Farms vary considerably in the standard of sanitation maintained. The question arises whether a farmer who conscientiously applies house fly control measures on his premises will find his efforts frustrated by invasions of house flies breeding on less sanitary farmsteads in the neighborhood. To answer this question it is necessary to understand the factors that affect house fly dispersal. These include wind direction, intensity of wind-borne odors and possibly variation in the tendency of flies to migrate. Some of these questions were answered in investigations during the summer of 1954, by releasing and recovering radioactive house flies in a dairy community near Fort Whyte, Manitoba.

Considerable study on the dispersion of house flies has been done by previous workers, including Parker (1916), Bishopp and Laake (1921), Lindquist (1951), Shoof *et al* (1952), Yates *et al* (1952), Quartermann *et al* (1954a and 1954b). Some workers claimed that house flies do orientate to wind-borne odors, others stated that wind has no effect on the dispersion of house flies, and still others believed that flies moved downwind in greater numbers than upwind. Considering the various locales and the different trapping methods employed, the conflicting results may be expected.

Methods

In this investigation, house fly dispersion was studied by recapturing radioactive flies which were previously released from a central point. The experimental area was a dairy community situated about two miles east of Fort Whyte, Manitoba. There were seven dairy barns and two swine barns located from one-quarter to about two miles away from the release points. Two releases of radioactive adult house flies were made in this area.

For the first release adult house flies were reared indoors. For the second, pupae were collected from manure piles and the adults emerged in a large screen cage in an insectary.

Three days prior to each release, food was removed and the flies were starved for two days. An aqueous solution of radioactive phosphorus (p^{32}) was then fed to the flies for one day. The amount of p^{32} used was approximately 20 microcuries per 1000 adult house flies. A check showed that 97 per cent of the flies were significantly radioactive according to the radioactivity monitor that was used. The machine was a portable Geiger counter with a thin end window tube. The window thickness was 1.5 milligrams per square centimetre.

The first release was made on August 5, 1954 in an open field in approximately the centre of the experimental area. The second release was made on one of the central farmsteads on August 23, 1954. On the day before the second release, fly samples were taken from the various barns and no radioactive flies were recovered from the first release.

1Based on part of a thesis submitted to the University of Manitoba, Department of Entomology, in partial fulfilment of the requirements leading to the degree of Master of Science.

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After the releases, fly samples were obtained by sweeping with a conventional insect net and by hanging sticky fly-papers in the heavily fly infested parts of the barns.

Results and Discussion

First Release

The mean daily temperature during the three day collecting period was 65.8°F. The mean maximum was 74.°F. and the mean minimum was 56.8°F. During the day of the release the wind blew at an average velocity of eight miles per hour from the northeast. During the second and third collecting days the wind direction was inconsistent and changed frequently.

Of the 15,000 radioactive adult house flies released, 548 or 3.7 per cent were recovered during the three collecting days. About 23,000 house flies were checked for radioactivity. Five hours after the release, 280 or 51.1 per cent of the tagged flies eventually recovered were caught, some of them two miles from the release point. During the evening of the day of the first release, 221 or 79 per cent of the tagged flies caught that day were collected on two farms that lay directly upwind from the release point. During the three day collecting period, 65 per cent of the recovered radioactive flies were caught on these two farms.

The uneven distribution of radioactive flies suggests that the released flies did not disperse at random from the release point but orientated to the wind or wind-borne odors. Some workers, Parker (1916), Bishopp and Laake (1921) and Lindquist (1951), suggested that there is little or no relation between the wind direction and fly dispersion. This discrepancy may have been the result of the different trapping methods used. There workers used small baited traps whereas in this investigation the entire farm yard served as a bait and was undoubtedly a more effective attractant over a greater distance. The small quantity of bait used in traps might not affect orientation of flies at a distant release point. These traps would then attract only those flies that chance to arrive in their vicinity. It is understandable that under these circumstances house flies might disperse at random. Odor is the primary stimulus affecting orientation of house flies and wind acts as a vehicle for the odor.

Second Release

In the second release an attempt was made to determine whether house flies disperse from one farm unit to another within the experimental area.

During the 17 day collecting period following the second release, the mean temperature was 60.5°F., the mean maximum 70.8°F., and the mean minimum was 50.2°F. Heavy rains, drizzle, cloud cover and prevailing cool north winds occurred during this period.

Of the 10,000 tagged adult house flies released, 7.17 per cent were recovered and about 125,000 flies were checked for radioactivity. About 79 per cent of the recovered radioactive flies were caught on the dairy farm on which they were released. The remaining 21 per cent were recovered on the other eight farms situated from one-half to two miles from the release point. Most of the tagged flies that left the release point were caught at two piggeries and two dairy barns nearest the piggeries. These four farms were close together and situated in the same direction from the release point.

Summary and Conclusions

Two releases of radioactive adult house flies were made in a dairy community during the summer of 1954 to determine (a) the effect of wind direction and wind-borne odors on dispersion of house flies; and (b) whether house flies disperse from one farm unit to another within the experimental area.

Adult house flies were tagged by feeding on radioactive phosphorus in aqueous solution sweetened with sucrose.

The results from the two releases indicate that (a) adult house flies orientate to wind-borne odors from farmyards, and (b) adult house flies migrate from one farmstead to another in appreciable numbers even in weather suboptimal for flight. This indicates the necessity of community rather than individual attempts to control this pest.

Acknowledgments

I would like to express my gratitude to Dr. A. J. Thorsteinson, Department of Entomology, the University of Manitoba, for his valued criticism during the investigations and in the preparation of this manuscript.

References

Bishopp, F. C. and E. W. Laake. 1921. Dispersion of house flies by flight. *Jour. Agr. Res.* 2(10): 729-66.

Hoffman, R. A. and A. W. Lindquist. 1951. Studies on treatment of flies with radioactive phosphorus. *Jour. Econ. Ent.* 44: 471-2.

Lindquist, A. W., W. W. Yates and R. A. Hoffman. 1951. Studies of the flight habits of three species of flies tagged with radioactive phosphorus. *Jour. Econ. Ent.* 44(3): 397-400.

Parker, R. R. 1916. Dispersion of *Musca domestica* L. under city conditions in Montana. *Jour. Econ. Ent.* 9(3): 325-54.

Quartermann, K. D., W. Mathis, and J. W. Kilpatrick. 1954a. Urban fly dispersal in the area of Savannah, Georgia. *Jour. Econ. Ent.* 48(3): 405-13.

Quartermann, K. D., J. W. Kilpatrick, and W. Mathis. 1954b. Fly dispersal in a rural area near Savannah, Georgia. *Jour. Econ. Ent.* 47(3): 413-19.

Shoof, H. F., K. E. Silverly, and J. A. Jensen. 1952. House fly dispersion in metropolitan areas. *Jour. Econ. Ent.* 45(4): 675-83.

Yates, W. W., A. W. Lindquist, and J. S. Butts. 1952. Further studies of dispersion of flies tagged with radioactive phosphoric acid. *Jour. Econ. Ent.* 45(3): 547-8.

Studies of Predators of the Balsam Woolly Aphid *Adelges piceae* (Ratz.) (Homoptera: Adelgidae)

I. Field Identification of *Neoleucopis obscura* (Hal.), *Leucopina americana* (Mall.) and *Cremifania nigrocellulata* Cz. (Diptera, Chamaemyiidae)[1,2]

By N. R. Brown[3] and R. C. Clark[4]

The introduction of predators of the balsam woolly aphid, *Adelges piceae* (Ratz.), from Europe to the Atlantic Provinces of Canada was commenced in 1933 as a joint project of the Forest Biology and Entomology Divisions of Science Service and the Commonwealth Institute of Biological Control. Several species were released between 1933 and 1941 but only one, *Neoleucopis obscura* (Hal.), became established and its control value was insufficient (Balch, 1952). The project was continued in 1951 and several additional species have been established. Studies of these predators and their place in the control complex in Canada were undertaken and this paper is the first in a series on the subject. It results from the necessity of rapid field identification of the various stages of all predators found. It deals with three closely related dipterous predators, two of which

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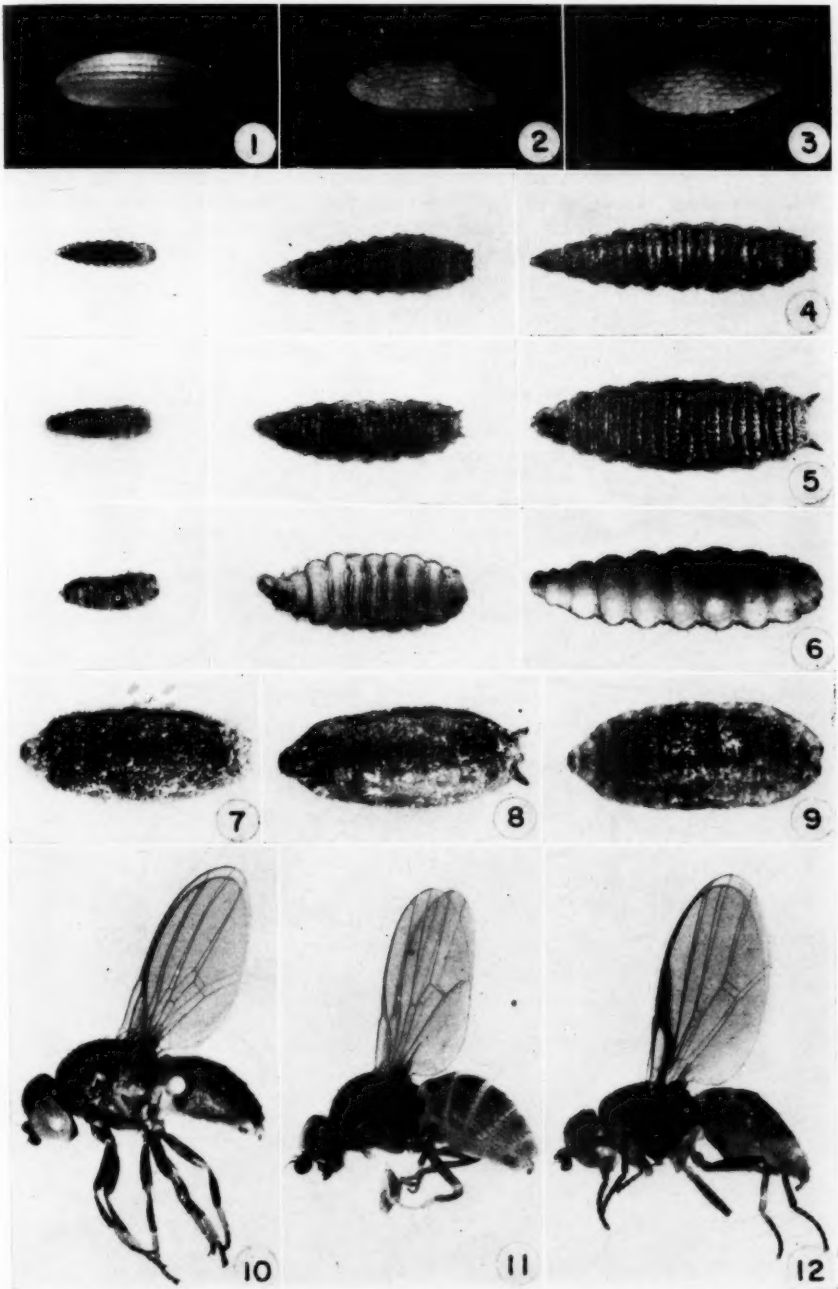
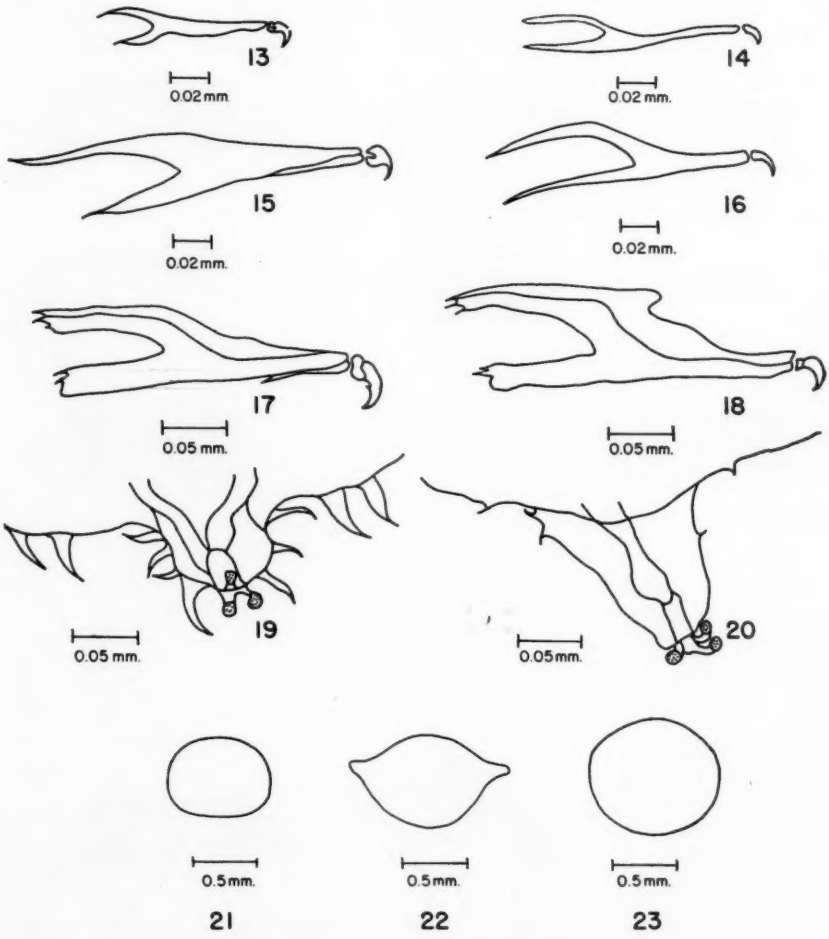


Figs. 1-12—1, *N. obscura*—egg (courtesy W. A. Reeks); 2, *L. americana*—egg; 3, *C. nigrocellulata*—egg; 4, *N. obscura*—first-, second-, and third-instar larvae; 5, *L. americana*—first-, second-, and third-instar larvae; 6, *C. nigrocellulata*—first-, second-, and third-instar larvae; 7, *N. obscura*—puparium; 8, *L. americana*—puparium; 9, *C. nigrocellulata*—puparium; 10, *N. obscura*—adult; 11, *L. americana*—adult; 12, *C. nigrocellulata*—adult.

were introduced, *N. obscura* and *Cremifania nigrocellulata* Cz., the other, *Leucopina americana* (Mall.), being native to Canada. Similar descriptions of the other species with a key will be published later.

Several authors have described one or more of the various stages of *N. obscura*. The adult was described briefly by Haliday (1833). Hardy (1871-72) gave a short description of the "maggot" and the "pupa-case" and a detailed description of the adult. Trägårdh (1931) described and illustrated the mature larva (including the spiracles and buccopharyngeal armature), the puparium, and a few parts of the adult, but none of the early larval instars. Atwood (1933) gave a very brief description of the larva, puparium, and adult in an unpublished



Figs. 13-23—Buccopharyngeal armature. *N. obscura*—13, First-instar larva; 15, Second-instar larva; 17, Third-instar larva. Buccopharyngeal armature. *L. americana*—14, First-instar larva; 16, Second-instar larva; 18, Third-instar larva. Posterior respiratory processes, third-instar larvae—19, *N. obscura*; 20, *L. americana*. Cross-section of puparia—21, *N. obscura*; 22, *C. nigrocellulata*; 23, *L. americana*.

	Neoleucopis obscura (Hal.)	*Leucopina americana* (Mall.)	*Cremifania nigrocellulata* Cz.
Egg	(See Fig. 1)	(See Fig. 2)	(See Fig. 3)
Length	Av. 0.42 mm. Range 0.29–0.47.	Av. 0.43 mm. Range 0.43–0.44.	Av. 0.420 mm. Range 0.414–0.432 (3).
Width	Av. 0.17 mm. Range 0.15–0.18.	Av. 0.16 mm. Range 0.15–0.17.	Av. 0.156 mm. Range 0.144–0.180 (3).
Colour	Silvery grey.	Creamy-white.	Intense white.
Shape	Elongate-oval, tapering more gradually at cephalic pole. More strongly arched on dorsal than ventral side.	Elongate-oval, tapering more gradually at cephalic pole. Slightly arched dorsally. Venter almost flat.	Elongate-oval, slightly tapered toward the cephalic pole. Circular in cross-section.
Surface	Dorsum striate longitudinally. Venter mostly glabrous.	Longitudinal striations slightly wavy; present over entire egg.	Covered with small oval depressions arranged in longitudinal lines, alternating in adjacent rows in a brick-like pattern.
Position	Deposited on bark; firmly attached.	Deposited loosely on or in masses of *Adelges* "wool".	Deposited on bark beside host egg masses; not firmly attached.
Larva, First Instar	(See Fig. 4)	(See Fig. 5)	(See Fig. 6)
Body			
Length	Av. 0.76 mm. Range 0.50–1.04 (12).	Av. 1.26 mm. Range 0.39–1.82 (11).	Av. 0.477 mm. Range 0.450–0.504 (2).
Width	Av. 0.22 mm. Range 0.17–0.25 (12).	Av. 0.48 mm. Range 0.15–0.62 (11).	Av. 0.153 mm. Range 0.144–0.162 (2).
Buccopharyngeal armature	(See Fig. 13)	(See Fig. 14)	(Not studied; see Delucchi and Pschorn-Walcher, 1954.)
Length of ventral arm	Av. 0.081 mm. Range 0.075–0.093 (3).	0.157 mm. (1).	
Length of terminal hook	Av. 0.0128 mm. Range 0.0120–0.0132 (3).	0.0189 mm. (1).	
Colour	Dirty yellowish-grey or whitish.	Creamy-white.	Grey-white, almost transparent on hatching.
Shape			
Body	Spindle-shaped.	Wedge-shaped	Almost oval, slightly tapered toward anterior end.
Anterior end	Tapered.	Tapered.	Broadly rounded.
Posterior end	Gradually tapered; rounded apically.	Blunt, not rounded apically.	Very broadly rounded.
Dorsum	Slightly arched.	Slightly arched.	Slightly arched. Lateral lobes more distinct.
Venter	Flat.	Flat.	Flat.
Larva, Second Instar	(See Fig. 4)	(See Fig. 5)	(See Fig. 6)
Body			
Length	Av. 1.77 mm. Range 1.04–2.08 (15).	Av. 2.07 mm. Range 1.62–2.44 (5).	Av. 1.783 mm. Range 1.557–2.202 (6).
Width	Av. 0.43 mm. Range 0.25–0.62 (15).	Av. 0.61 mm. Range 0.58–0.65 (5).	Av. 0.438 mm. Range 0.376–0.573 (6).
Buccopharyngeal armature	(See Fig. 15)	(See Fig. 16)	(Not studied; see Delucchi and Pschorn-Walcher, 1954)
Length of ventral arm	Av. 0.142 mm. Range 0.119–0.191 (32).	0.2025 mm. (1).	
Length of terminal hook	Av. 0.0185 mm. Range 0.0132–0.0220 (31).	0.0243 mm. (1).	
Colour	Yellow-grey.	Creamy-white.	Dirty yellow-grey. Dorsal rows of heavy, dark spines arranged on each side of the intersegmental folds sometimes discernible.
Shape			
Body	Spindle-shaped.	Wedge-shaped.	Wedge-shaped.
Anterior end	Strongly tapered.	Strongly tapered.	Strongly tapered.
Posterior end	Rounded apically; posterior respiratory processes very small, not visible to naked eye.	Blunt; posterior respiratory processes large, prominently visible to naked eye.	Rounded apically, posterior respiratory processes very small, visible as two tiny black dots, widely separated.
Dorsum	Slightly arched.	Arched.	Slightly arched; segmentation very distinct; lateral lobes prominent.
Venter	Flat.	Flat.	Rounded.

	Neoleucopis obscura (Hal.)	*Leucopina americana* (Mall.)	*Cremifania nigrocellulata* Cz.
Larva, Third Instar	(See Fig. 4)	(See Fig. 5)	(See Fig. 6)
Body			
Length	Av. 2.74 mm. Range 1.75–3.91 (66).	Av. 3.17 mm. Range 2.37–3.89 (21).	Av. 3.320 mm. Range 2.646–4.284 (24).
Width	Av. 0.68 mm. Range 0.46–0.93 (66).	Av. 0.89 mm. Range 0.65–1.14 (22).	Av. 0.960 mm. Range 0.756–1.134 (24).
Buccopharyngeal armature	(See Fig. 17)	(See Fig. 18)	(Not studied; see Delucchi and Pschorn-Walcher, 1954)
Length of ventral arm	Av. 0.233 mm. Range 0.180–0.268 (86).	Av. 0.2465 mm. Range 0.2268–0.2604 (13).	
Length of terminal hook	Av. 0.0292 mm. Range 0.0220–0.0396 (56).	Av. 0.0265 mm. Range 0.0252–0.0294 (13).	
Posterior respiratory processes	(See Fig. 19)	(See Fig. 20)	
Basal diameter	0.053 mm.	0.092 mm.	
Length	0.079 mm.	0.160 mm.	
Distance between bases	0.160 mm., or 2X length.	0.240 mm., or 1.5X length	
Length of terminal filaments	0.018 mm.	0.036 mm.	
Width of terminal filaments	0.009 mm.	0.008 mm.	
General appearance	Posterior respiratory processes inconspicuous under *Adelges* "wool" covering.	Posterior respiratory processes conspicuous, easily seen with naked eye.	Posterior respiratory processes very small, visible as two tiny black dots, widely separated.
Detailed description (living larva)			
Length	About 3.5 mm., full grown.	About 4.0 mm., full grown.	About 4.0 mm., full grown.
Width	About 1.0 mm., full grown.	Over 1.0 mm., full grown.	Over 1.0 mm., full grown.
Colour	Dirty yellowish-grey or whitish.	Dull greyish or whitish.	Dirty grey; newly-moulted larvae yellowish.
Shape			
Body	Spindle-shaped	Elongate-oval.	Wedge-shaped.
Anterior end	Strongly tapered.	Strongly tapered.	Gradually tapered from mid-section.
Posterior end	Gradually tapered, rounded apically.	Weakly tapered, rounded apically.	Weakly tapered, rounded apically.
Dorsum	Somewhat arched.	Strongly arched.	Strongly arched.
Venter	Flat.	More or less rounded (more nearly circular in cross-section than *N. obscura*).	Rounded; in cross-section larva is almost oval.
Segmentation	Very distinct; segments with annular ridged rings. Lateral lobes prominent.	Indistinct, without strong dark segmentation lines as in *N. obscura*. Lateral lobes indistinct.	Very distinct, lateral lobes very prominent.
Texture of integument	Very rough, granular; abdomen covered with short cuticular spines.	Smooth, as compared with *N. obscura*.	Smooth; colourless cuticular processes not visible with hand lens; dorsal rows of heavy, dark spines arranged on each side of intersegmental folds sometimes discernible.
Covering	Dorsum usually covered with "wool", has a powdery appearance.	Naked; not covered with "wool".	Naked; not covered with "wool".
General description for field identification	Dirty grey or whitish; strong segmentation lines; posterior respiratory processes not visible; granular appearance, covered with small pieces of "wool"; movement undulating.	Dull greyish; segmentation lines weak; posterior respiratory processes visible with naked eye; body smooth; not covered with "wool"; movement undulating.	Dirty grey; segmentation distinct with prominent lateral lobes; posterior respiratory processes distinct, appearing as small black dots; body smooth, not covered with "wool"; movement looping, geometrid-like.

report. Wilson (1938) provided a few notes on the egg, larva, and puparium. Reeks (1943) included a short description of the egg and photographs of the egg and the buccopharyngeal armature of the second- and third-instar larvae in an unpublished report.

Malloch (1921) published a good description of the adult of *L. americana* but there are no descriptions of the immature stages.

Only one paper on *C. nigrocellulata* (Delucchi and Pschorn-Walcher, 1954) was found in a survey of the literature. This gives information on the mor-

	Neoleucopis obscura (Hal.)	*Leucopina americana* (Mall.)	*Cremifania nigrocellulata* Cz.
Puparium	(See Fig. 7)	(See Fig. 8)	(See Fig. 9)
Detailed description			
Length	Av. 2.0 mm. Range 1.8–2.3.	Av. 2.7 mm. Range 2.1–2.9 (including respiratory processes). Av. 2.6 mm. Range 2.0–2.8 (excluding respiratory processes).	Av. 2.881 mm. Range 2.583–3.213.
Width	Av. 0.9 mm. Range 0.8–1.0.	Av. 1.05 mm. Range 0.9–1.1.	Av. 1.210 mm. Range 1.134–1.386.
Colour	Yellow-brown or dark brown.	Reddish-brown.	Dark brown.
Shape			
From above	Oval, sides weakly rounded.	Oval, sides more strongly rounded.	Elliptical to hypocycloidal.
Cross section	Approximately semi-circular (see Fig. 21).	Almost circular (see Fig. 23).	Triangular (see Fig. 22).
Anterior end	Somewhat contracted, upper surface depressed and flattened.	Strongly contracted and flattened dorso-ventrally so that anterior end projects like a small knob.	Concave dorsally, pointed.
Posterior end	Broadly rounded.	Broadly rounded.	Broadly rounded; small median notch results in bilobed appearance.
Dorsum	Arched.	Very strongly arched.	Median part weakly arched; strongly recurved lateral carinae present.
Venter	Flat.	Slightly less arched than dorsum; very little flattened.	Blunt wedge-shaped (semi-hexagonal).
Segmentation	Usually fairly distinct as dark lines.	Generally indistinct; evident depression along each side.	Segmentation lines faintly visible; lateral lobes prominent.
Texture of integument	Roughened.	Usually smooth, sometimes covered with very fine white dusting.	Granular.
Covering	Generally more or less completely covered with small pieces of "wool".	Usually no "wool", surface smooth.	Not covered with "wool".
Posterior respiratory processes	Inconspicuous, usually hidden by adhering "wool".	Very long, conspicuous, projecting postero-dorsally, clearly visible with naked eye; diverging toward apices.	Visible as small black dots, widely separated.
Position	Generally on bark surface; occasionally in soil.	Generally on bark surface; occasionally in soil.	Usually in soil, occasionally on bark surface.
General description for field identification	Yellow or dark brown; flattened ventrally; usually covered with small pieces of "wool"; posterior respiratory processes small, not visible with naked eye, segmentation generally distinct.	Reddish-brown; almost circular in cross-section; robust, larger on average than *N. obscura;* usually smooth and not covered with "wool"; posterior respiratory processes very long, conspicuous, easily visible with naked eye; segmentation generally indistinct.	Dark brown; triangular in cross-section with strong recurved lateral carinae; naked, no "wool" covering; surface granular; posterior respiratory processes visible as small black dots; segmentation lines distinct; lateral lobes prominent.

	Neoleucopis obscura (Hal.)	*Leucopina americana* (Mall.)	*Cremifania nigrocellulata* Cz.
Adult	(See Fig. 10)	(See Fig. 11)	(See Fig. 12)
General Colour	Black, usually dull, sometimes somewhat shining.	Light grey, dull pubescent-like.	Dull grey.
Front of head	Uniform dull black.	Dull grey, median dorsal light vertical line; vertical light grey line along median side of each eye; antennae set in grey round area.	
Thorax	Black.	Grey, usually with two distinct brownish longitudinal dorso-lateral lines, one on each side.	Dorsum dark brown; pleura and venter light brown.
Halteres	Yellow, club round, very conspicuous.	Whitish or flesh-coloured; longer than in *N. obscura*.	Light grey or flesh-coloured.
Legs	Black, except basal tarsal segments.	Femora grey; tibiae dull grey-brown; tarsi yellowish, becoming dark toward apices.	Brown, with femora darker on dorsal side; prothoracic legs darker than others, same colour as dorsum of thorax.
Wings	Clear.	Clear.	Stigma dark brown; typically holds wings out from body, at slight upward angle and vibrating.
Abdomen	Dorsum black, venter mostly light straw-coloured; callow adults with much more extensive light areas.	Very light grey; powdered appearance.	Dorsum dark grey-brown, venter light grey.

phology and biology in Europe. The morphological descriptions as given are not suitable for field identification and separation from closely related species.

Larval measurements were made from individuals preserved in 70 per cent ethyl alcohol. Measurements and illustrations of larval mouth parts were made from material cleared in KOH and mounted on microscope slides in Canada balsam. Positive identification of species involved was made by rearing larvae through to the adult stage. Adults were identified by G. E. Shewell and J. F. McAlpine of the Unit of Systematic Entomology and Biological Control, Entomology Division, Ottawa. Photographic work was done by E. B. Bates, Forest Biology Laboratory, Fredericton and the Bio-Graphic Unit, Science Service, Ottawa.

The foregoing tabulations are of characters mostly useful for field identification and visible with the aid of a hand lens of 6 to 10 power. Also included are some characters discernible only by microscopic study, particularly those of larval buccopharyngeal armature.

References

Atwood, C. E. 1933. Control (of *Adelges piceae*) by predators. *In* Annual Report, Division of Forest Insects, Fredericton, N.B. (Unpublished).

Balch, R. E. 1952. Studies of the balsam woolly aphid, *Adelges piceae* (Ratz.), and its effects on balsam fir, *Abies balsamea* (L.) Mill. Canada, Dept. Agr. Pub. 867.

Delucchi, V. and H. Pschorn-Walcher. 1954. *Cremifania nigrocellulata* Czerny (Diptera, ? Chamaemyiidae), ein Raüber an *Dreyfusia* (*Adelges*) *piceae* Ratz. (Hemiptera, Adelgidae). *Zeitschr. f. angew. Entomologie* 36 (1): 87-107.

Haliday, A. H. 1833. Article XVII. Catalogue of Diptera occurring about Holywood in Downshire. *The Entomological Magazine* 1: 173.

Hardy, J. 1871-72. Memoirs on Scottish Diptera. *Leucopis obscura. Scottish Naturalist* 1: 256-258.

Malloch, J. R. 1921. Article XIV. Forest insects in Illinois. I. The subfamily Ochthiphilinae (Diptera, Family Agromyzidae). *Bull. Illinois State Nat. Hist. Survey* 13: 354-355.

Reeks, W. A. 1943. *Leucopis obscura. In* Annual Report, Dominion Entomological Laboratory, Fredericton, N.B. (Unpublished).

Trägårdh, I. 1931. Zwei forstentomologisch wichtige Fliegen. *Zeitschr. f. angew. Entomologie* 18 (3): 672-690.

Wilson, F. 1938. Notes on the insect enemies of *Chermes* with particular reference to *Pineus pini* Koch and *P. strobi* Hartig. *Bull. Ent. Res.* 29: 373-389.

Colorimetric Estimation of Dyed Insecticide Spray Deposit Using a Paper Sampling Surface[1]

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During spray trials carried out recently for European pine shoot moth control in Ontario, a method was developed to assess dyed, oil-based insecticide spray deposits colorimetrically using a paper sampling surface. The procedure permits intensive sampling without the necessity of handling fragile or bulky sampling kits in the field.

The sampling material consists of a disc of filter paper, which must be of sufficient size to give adequate colour when extracted. Filter paper was chosen because of its absorptive nature, uniformity, and freedom from coloured materials. Whatman No. 1, 9.0cm. in diameter, was found to constitute a satisfactory sampling surface at the dosages employed in the experimental spraying operation (up to 2 gallons per acre of a dyed oil-based 10 per cent DDT solution[2]). The paper was stapled to a flat cardboard surface and the sampling units were distributed over the area to be sprayed. After the spraying operation, the papers were removed from the cardboard backing and stacked loosely. No cross-contamination occurred at the dosages encountered. Field sample collections were made as rapidly as possible after spraying to minimize fading of the dye in sunlight. To extract the dye, each filter paper was folded longitudinally and inserted into a 12.5 mm. colorimeter tube. Ten millilitres of reagent-grade benzene were added and the tube corked. The tube was shaken for a few seconds to dissolve the dye. The paper was then removed from the tube and a colorimetric reading taken on the solution, using a Klett-Summerson colorimeter.

To obtain a standard colour curve a solution of known concentration of dyed spray mix in benzene was prepared. Aliquots of this solution were taken by means of a micropipette. Each aliquot was run over the surface of a blank filter paper, taking care that the paper retained all the solution. The benzene solvent was allowed to evaporate, leaving the dyed insecticide on the paper. Each paper was folded and extracted by the procedure described above. The concentration of insecticide was then plotted against the colorimeter reading. Ten millilitres of benzene shaken with a filter paper containing no coloured material served as a colorimeter blank.

To obtain information on whether retention of dye by filter paper in benzene solution occurred, and if so, to what degree, the filter-paper method was compared with one using glass plates to collect the spray deposit. The results indicate that spray deposits sampled by the filter-paper method give dyed solutions having slightly lower optical densities than deposits sampled by the glass-plate method. The density difference is estimated to be of the order of 1 to 2 per cent, and is interpreted as being due to retention of a small amount of dye by the paper itself.

1Contribution No. 296, Forest Biology Division, Science Service, Department of Agriculture, Ottawa, Canada.

2Twenty-five per cent emulsifiable DDT concentrate diluted to 10 per cent with No. 2 fuel oil. The solution was dyed with 0.5 per cent of Du Pont Oil red dye and filtered.

The Reaction of Certain Lepidopterous and Hymenopterous Larvae to *Bacillus sotto Toxin*[1]

T. A. Angus[2]

Introduction

Bacillus sotto Ishiwata is a bacterial pathogen of the larvae of *Bombyx mori* L. (the silkworm) and it has been shown (2) that the toxin it produces is associated with the crystalline inclusions (5) found in sporulated cells of this microorganism. The pathogenicity of *B. sotto* for the larvae of certain insect species and the properties of *B. sotto* toxin have been described previously (1, 2, 3). In the present paper the effect of the toxin on the larvae of several insect species is reported.

Methods

The preparation of *B. sotto* toxin and the methods of testing its toxicity have been described (1, 3). Briefly, the dried toxin was dissolved in 0.01 N sodium hydroxide and applied with Methocel to the surface of leaves, which were then air dried and fed to larvae. In some cases a method of forced-feeding (1) was used. All larvae were starved for at least eight hours before being fed treated foliage. Control larvae were fed foliage coated with solvent and Methocel.

Results

The species tested and their susceptibility to the action of *B. sotto* toxin are shown in Table I, which also includes the pH of the mid-gut contents of each species.

The symptoms observed in silkworm larvae following ingestion of *B. sotto* toxin were identical with those caused by the whole organism (spores, crystals, and debris) with the exception that melanosis (1) sometimes was not apparent until after 36 hours. In insects killed with the whole organism this generally appears within 24 hours. In none of the other species tested were the symptoms as striking as in silkworm (1).

Larvae of some insect species, unlike those of *B. mori*, are reluctant to feed on foliage coated with *B. sotto* toxin. Feeding would begin but soon cease and the larvae would eventually die, apparently of starvation. When a method of forced-feeding was adopted to overcome this reluctance to feed, it appeared that the insects were perhaps unable to feed after ingestion of toxin. After larvae in forced-feeding tests had ingested the drop of toxin solution deposited on their mouth parts, they were placed on clean foliage, and it was noted that feeding was light. Control larvae that had ingested only toxin solvent fed readily.

When the behaviour and symptoms of all species are compared, the following categories or groups can be distinguished:

Bombyx mori L., *Protoparce quinquemaculata* (Haw.) and *P. sexta* (Johan.) feed readily on foliage coated with toxin. Ingestion of toxin was followed by a profound irreversible paralysis within two to six hours and death in 24 hours. There was no regurgitation or diarrhea, but an oral and occasionally an anal discharge resulted. This was probably leakage from the gut, since muscular control was lacking (1).

Datana integerrima G. & R., *D. ministra* (Drury), and *Nymphalis antiopa* (L.) feed readily on foliage coated with toxin. Ingestion of toxin was followed by

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TABLE I
Susceptibility of larvae of several insects to *B. sotto* toxin

Insect	pH of mid-gut contents*	Susceptibility
LEPIDOPTERA		
Bombyx mori L.	10.4	
Protoparce quinquemaculata (Haw.)	9.9	These species are susceptible. Paralysis followed by death in 24 hours. They feed readily on coated foliage.
Protoparce sexta (Johan.)	9.6	
Datana integerrima G. & R.	9.9	
Datana ministra (Drury)	9.9	
Nymphalis antiopa (L.)	10.2	
Anisota rubicunda (F.)	9.4	These species are susceptible. No paralysis is seen. They feed poorly on coated foliage.
Anisota senatoria (A. & S.)	9.9	
Porthetria dispar L.	9.4	
Choristoneura fumiferana (Clem.)	9.2	
Cirphis unipuncta (Haw.)	—	
Galleria mellonella (L.)	8.4	
HYMPENOPTERA		
Diprion hercyniae (Htg.)	8.5	These species are not susceptible. They feed readily on coated foliage.
Neodiprion americanus banksianae (Roh.)	8.1	
Neodiprion abietis (Harr.)	8.5	
Neodiprion sertifer (Geoff.)	8.2	

*maximum of mid-gut contents as given by Heimpel (6, 7 or in personal communications).

partial paralysis or sluggishness, usually within six to twelve hours but sometimes earlier, and death followed in 24 hours. No true regurgitation or diarrhea was observed but leakage occurred.

Anisota rubicunda (F.), *A. senatoria* (A. & S.), and *Porthetria dispar* L. began feeding on foliage coated with toxin but soon ceased. The insects became sluggish, but not paralyzed. Death did not occur until after 36 hours, and with *P. dispar* and *A. senatoria*, sometimes not before 100 hours. If, after 24 hours, the coated foliage was replaced with clean foliage, in most cases no feeding or almost none took place. If larvae of these species were forced to ingest toxin and were then placed on clean foliage, some feeding took place but soon ceased and the insects died within 36 to 72 hours. It is thought that the toxin interferes with the desire or ability of insects to feed. Since normal larvae can survive for more than 72 hours without food death can not be due simply to starvation but there must be some additional toxic effect which is not at present understood.

The other species tested (Table I) were unaffected by ingestion of *B. sotto* toxin, although they fed readily on coated foliage.

Discussion

Heimpel (6) in a recent study of *Bacillus cereus* strain PR-1017, pathogenic for larch sawfly, has shown that the gut pH is an important factor in insect diseases. The PR-1017 strain, which does not produce crystalline inclusions, is believed to be pathogenic because it produces the enzyme phospholipase. If this organism is fed to an insect, such as the silkworm, with a gut pH beyond 9.0, multiplication of vegetative cells is restricted and production of the toxic enzyme is limited. Even if the enzyme were produced, the gut pH is beyond its optimum activity range (6). The larch sawfly, which is killed by the PR-1017 strain, has a midgut pH between 7.1 and 8.5. When a sporulated culture of *B. sotto* is fed to larch sawfly or other sawfly larvae (see Table I), the insects are

not paralyzed, presumably because the toxin-containing crystals are not soluble at the pH of the sawflies' gut. All the susceptible species have relatively alkaline midgut contents (pH 9.0-10.5), and this suggests that there is a causal relationship between gut pH and susceptibility.

A widely-accepted explanation of the mode of action of a bacterial disease of a lepidopterous insect has been given by Mattes (see Steinhaus (8)). He described the course of infection in larvae of *Ephestia kühniella* Zell. fed flour contaminated with spores of *Bacillus thuringiensis* Berliner as: "1) ingestion of the spores and their germination in the midgut of the insect; 2) formation of large numbers of vegetative cells in the gut; 3) damaging the gut epithelium by the enzymatic activity of the bacillus as it flourishes in the gut contents; 4) migration of the bacteria between the host's cells into the body cavity (according to Berliner (1915) the bacteria enter the body cavity through openings in the gut wall that result from the disintegrating process); 5) abundant growth of the bacillus in the hemolymph; 6) disintegration of the internal tissues (e.g. the fat body) as a result of bacterial enzymes; 7) appearance of the external symptoms of the disease; 8) penetration of the bacteria into tissues of the nervous system, resulting in the death of the larva; 9) further disintegration of all tissues and the drying or mummification of the larval remains; 10) formation of spores by the bacteria."

The mode of action of *B. sotto* in silkworm larvae is not known but recent observations (4) suggest that Mattes' description does not apply. Paralysis in silkworm larvae treated with *B. sotto* (either whole organism or toxin) is accompanied by an increase in the alkalinity of the blood and a decrease in alkalinity of the gut (4). A provisional scheme (with question marks indicating areas of doubt) for the *B. sotto* disease in silkworm larvae might take the following form: 1) ingestion of the spores and crystal; 2) the spores do not germinate but the crystals or some part of them dissolve when acted on by the gut juice; 3)(?) the released toxin acts at the sensitive site, possibly the gut wall; 4) the haemolymph increases in alkalinity; 5) paralysis coincides with maximum alkalinity of haemolymph; 6) death of larvae; 7)(?) germination of the spores in the gut and saprophytic growth of the organism in the larval tissues.

It has been suggested that the germination of ingested spores is inhibited by the alkalinity of the midgut contents, thus the decrease in alkalinity of the gut contents of paralyzed larvae might be sufficient to permit germination of the spores before death. This may be the explanation of conditions that Toumanoff and Vago (9) described as intermediate between toxemia and septicemia.

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References

1. Angus, T. A. General characteristics of certain insect pathogens related to *Bacillus cereus*. [In press. Can. J. Microbiol. 2: No. 2.] To be completed.
2. Angus, T. A. Association of toxicity with protein-crystalline inclusions of *Bacillus sotto* Ishiwata. [In press. Can. J. Microbiol. 2: No. 2.] To be completed.
3. Angus, T. A. Extraction, purification, and properties of *Bacillus sotto* toxin. [In press. Can. J. Microbiol. 2: No. 2.] To be completed.
4. Angus, T. A. and Heimpel, A. M. 1956. An effect of *Bacillus sotto* on the larvae of *Bombyx mori*. *Can. Entomologist* 88(3).

5. Hannay, C. L. Crystalline inclusions in aerobic spore forming bacteria. Nature 172: 1004. 1953.
6. Heimpel, A. M. The pH in the gut and blood of the larch sawfly, *Pristiphora erichsonii* (Htg.) and other insects with reference to the pathogenicity of *Bacillus cereus* Fr. and Fr. Can. J. Zool. 33: 99-106. 1955.
7. Heimpel, A. M. Further observations on the pH in the gut and the blood of Canadian forest insects. In press. Can. J. Zool.
8. Steinhaus, E. A. Possible use of *B. thuringiensis* Berliner as an aid in the biological control of the alfalfa caterpillar. Hilgardia 20: 359-381. 1951.
9. Toumanoff, C. and Vago, C. La nature de l'affection des vers à soie due à *Bacillus cereus* var. *alesti* et les modalités d'action de ce bacille. Ann. inst. Pasteur 83: 634-639. 1952.

Book Review

THE PHYSIOLOGY OF DIAPAUSE IN ARTHROPODS. By A. D. Lees, pp. x and 151, 1955. Published in Canada by The Macmillan Company of Canada, Ltd. Price $2.15.

This little book is No. 4 in the series of Cambridge Monographs in Experimental Biology. The author is a member of the Agricultural Research Council's Unit of Insect Physiology, well-known for the many contributions to science made by its members.

In his Introduction Dr. Lees outlines the history of the word diapause from its embryological beginnings with Wheeler in 1893. Then, using Shelford's (1929) definition of diapause as a 'spontaneous' arrest of development or activity, Dr. Lees embarks on a brief discussion of the distinction between diapause and quiescence. He finds Shelford's definition "no longer wholly appropriate" as the cause of onset of diapause can often be traced back to the action of the environment. Surely, even with the benefit of quotation marks the word 'spontaneous' is an anachronism in modern biology. However, Dr. Lees apparently considers that diapause may be said to be 'spontaneous' in that it may eventually become independent of the primary response. He believes that the terms diapause and quiescence may be retained in entomological usage as there is a valid basis by which they may be distinguished. He considers that dormancy that is directly responsive to an environmental factor is quiescence, and that dormancy that is only remotely responsive to an environmental factor is diapause. However, he admits that this distinction cannot be pressed too far when environmental factors other than temperature are operative.

In the 140 pages that follow this brief introduction Dr. Lees discusses, with copious references to world literature and a welcome familiarity with Canadian contributions to the subject, such aspects as environment and the onset and termination of diapause, the role of water in the arrest of growth, metabolic adjustment and humoral control. He also presents chapters on the inheritance of diapause, diapause in parasitic insects and diapause and phenology. These pages are crammed with information—fascinating and thought-provoking. Here are some examples. Is diapause a pathological condition concurrent with low metabolism (p. 116)? Does a hormone terminating diapause synthesize certain vitamins (p. 117)? Do the neuro-secretary cells of the brain retain a sensory function and form a link between environment and the endocrine system (p. 112)? Are insects with obligatory diapause indifferent to the environment (p. 40)? Do the properties of a host insect's blood affect the diapause of its parasite (p. 83)? May the intensity of diapause vary in different parts of an insect's range, say between Gaspé and New Brunswick (p. 60)?

But after all this the author's ten concluding lines seem very deflating. A final co-ordinating chapter would surely be much appreciated and in subsequent editions Dr. Lees may perhaps vouchsafe us one. Again, while Dr. Lees, grammar is, as expected, impeccable and his avoidance of 'jargon' (Quiller-Couch) almost complete, the complexity of his style at times delays the reader, perhaps as a result of compression of documented material, e.g.: ". . . . the immediacy of the response to the environmental factor remains a valid basis for distinction, particularly if this factor is temperature".

But perhaps this is mere carping.

The phenomenon of diapause interests all entomologists in varying degrees and is of practical importance to those who rear and try to control insects. Physiologists and ecologists not primarily concerned with insects will also be interested in diapause. Thus Dr. Lees' scholarly, well-documented little book. incorporating what must be almost all that is known on the subject, should surely receive a warm welcome among entomologists and may well become required reading in many a graduate course. The book is enlivened with graphs, tables and figures; complemented by a list of references, detailed, neat and clear; and completed by a short index.

Dr. Lees is to be congratulated. Most of us would be glad to have written such a monograph. At its Canadian list price it is one of to-day's book bargains.

A. J. MUSGRAVE.

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